

EFFECTS OF SUPPLEMENTATION WITH CREATINE MONOHYDRATE AND BETA-ALANINE, ALONE OR COMBINED, ON REPEATED SPRINT PERFORMANCE AND PHYSIOLOGICAL PARAMETERS IN AMATEUR TEAM AND RACKET SPORT PLAYERS

Anne Delextrat¹, Nese Targen², Georgina Impson-Davey¹, Daniel Kapsis¹, James Bateman³, Nicolas Terrados⁴, and Julio Calleja-González⁵

¹*Sport and Health Science Department, Oxford Brookes University, UK*

²*Nutrition and Dietetics Department, Maidstone and Tunbridge Wells NHS Trust, UK*

³*University College Birmingham, Birmingham, West Midlands, UK*

⁴*Regional Sports Medicine Unit of Asturias, Aviles Municipal Sports Foundation and Department of Functional Biology, University of Oviedo, Oviedo, Spain*

⁵*Laboratory of Analysis of Sport Performance, Sport and Physical Education Department, University of the Basque Country, Vitoria-Gasteiz, Spain*

Original scientific article

DOI: 10.26582/k.52.1.15

Abstract:

The purpose of this study was to compare the combined effects of creatine monohydrate (Cr) and beta-alanine (BA) with their isolated use on performance and physiological parameters during repeated sprint sequences (RSS). Forty-four male (n=34) and female (n=10) amateur team- and racket sport players (25.1±3.1 years; 175.2±9.8 cm; 76.0±10.3 kg; 15.2±6.8% body fat) performed 10 repetitions of 6-s sprints with departure every 30 s, before and after a 28-day supplementation period with either Cr (n=11; 5 g·day⁻¹), BA (n=10; 6 g·day⁻¹), combined Cr and BA (n=12; 5 g·day⁻¹ of Cr plus 6g·day⁻¹ of BA) or placebo (n=11; 11 g·day⁻¹ of rice flour). Peak (PP) and mean power (MP), performance decrement (%Dec), heart rate (HR), blood lactate concentration (LA), and perceived exertion (RPE) were measured. Analyses of variance (ANOVA) were used to determine the effects of groups (Cr, BA, CrBA, P), sprint number (1 to 10), and time (pre- vs. post-supplementation) on all variables. A significant increase in PP was shown in the post- compared to the pre-supplementation in Cr (+5.2%) and BA (+5.2%) groups only (p<.05), and significant decreases in MP in all groups (3.7% to 6.4%, p<.05), except BA. %Dec was significantly decreased post-supplementation in the Cr group only (17.4%, p<.05). No effects were shown on HR, RPE and LA (p<.05). These results show no additional benefits of the combination of Cr and BA on RSS performance and suggest that longer sprint or total exercise duration might be necessary to observe the benefits of the combined supplementation.

Key words: lactate, power, recovery, short sprints, repeated sprint sequence

Introduction

Throughout a team or racket sport match, players are required to perform short maximal repeated sprints (< 6 s), separated by brief recovery periods (20-25 s), during extended periods of time, defined as repeated sprint sequences (RSS) (Buchheit & Laursen, 2013; Girard, Mendez-Villanueva, & Bishop, 2011). However, maximal sprint performance during RSS cannot be maintained for a very long time, as fatigue has been shown to develop as early as after the first sprint (Mendez-Villanueva, Hamer, & Bishop, 2007). Multiple factors can

cause fatigue during RSS, including limitations in anaerobic energy supply, metabolite accumulation in muscles and neural factors (Girard, et al., 2011).

During RSS, intramuscular PCr stores act as a short-term energy supply (Harris, Soderlund, & Hultman, 1992). However, intramuscular PCr stores are limited and cannot be fully replenished during typical short recovery periods experienced in RSS (Gaitanos, Williams, Boobis, & Brooks, 1993; Spencer, Bishop, Dawson, & Goodman, 2005). Supplementation with creatine monohydrate (Cr) has been shown to increase intramuscular PCr

stores and increase the contribution of this source of energy to the total energy supply during RSS (Allen, Lamb, & Westerblad, 2008; Balsom, Soderlund, Sjodin, & Ekblom, 1995). Improvements in peak power and/or total mechanical work has been shown during RSS protocols including 6-12 sprint repetitions following the supplementation of Cr for six to 37 days, compared to a placebo (Kamber, et al., 1999; van Loon, et al., 2003; Yquel, Arsac, Thiaudiere, Canioni, & Manier, 2002; Ziegenfuss, et al., 2002).

The progressive depletion of PCr during repeated sprints activates the anaerobic glycolysis pathway (Gaitanos, et al., 1993), leading to the intramuscular accumulation of H⁺ if levels exceed removal via buffering. Muscle acidosis could have detrimental effects on muscle function, including inhibition of glycolytic enzymes, disruption in contractility and PCr resynthesis (Girard, et al., 2011; Spriet, Lindinger, McKelvie, Heigenhauser, & Jones, 1989). Beta-alanine (BA) is the rate-limiting substrate for the intramuscular production of carnosine, one of the main intramuscular buffers (Hobson, Saunders, Ball, Harris, & Sale, 2012). However, results on repeated efforts are equivocal with several studies showing no beneficial effect of four to five weeks following BA supplementation (6-6.4 g·day⁻¹) compared to a placebo on RSS performance indicators (Ducker, Dawson, & Wallman, 2013; Saunders, Sale, Harris, & Saunderson, 2010; Sweeney, Wright, Brice, & Doberstein, 2010), while significant improvements were shown with smaller doses but a similar supplementation duration on repeated knee extension strength (Derave, et al., 2007).

In view of the isolated benefits of Cr and BA, a few studies have considered their potential combined effects, showing significant benefits on endurance (Zoeller, Stout, O'Kroy, Torok, & Mielke, 2007) as well as single anaerobic performance (Hoffman, et al., 2006). Only two investigations have examined the combined effects of Cr and BA on long (30 s) repeated sprints, interspersed by 2-3-min of recovery (sprint interval training; SIT), showing contrasting results (Kresta, et al., 2014; Okudan, Belviranli, Pepe, & Gokbel, 2015). Okudan et al. (2015) showed significant improvements in mean power in the combined group, compared to pre-supplementation in sedentary men, while Kresta et al. (2014) did not show any additive benefits of BA and Cr compared to their single use in recreationally active women when participants pedaled as fast as possible prior to the application of the workload and sprint at all-out maximal capacity during the 30-s test with 3-min of passive rest in between. However, to our knowledge, there is no study on the effect of this combination during shorter repeated sprint events, such as RSS. Results from SIT cannot be applied to RSS, as it is well-

known that the contribution of different energy systems and limiting factors are different between these two types of intermittent exercises (Buchheit & Laursen, 2013; Girard, et al., 2011). For example, blood lactate values are usually greater during SIT than RSS, and the smaller recovery experienced during RSS compared to SIT might slow down the restoration of PCr stores or lactate clearance (Buchheit & Laursen, 2013).

Therefore, the main aim of this study was to compare the combined effects of Cr and BA with their isolated use on performance during RSS in team and racket sport players.

Methods

Participants

Fifty-two male (n=40) and female (n=12) amateur team and racket sport players were recruited from local clubs and university sport teams (Table 1). At the time of the study, they were involved in an average of 5-h of weekly training, including 2-h of resistance training and 3-h of aerobic-based exercise mixed with tactical work. Exclusion criteria were any known kidney or liver disease, lower limb injury, and use of nutritional supplements in the preceding four weeks. In addition, all participants were omnivores. They received detailed information about the study before giving informed written consent. Ethical approval for the study was granted by the university research ethics committee, in accordance with the Helsinki Board for the Protection of Human Subjects.

Design

This study used a randomized, double-blind, placebo-controlled, parallel group design.

Methodology

Procedures

All participants performed a familiarisation trial, and two main testing sessions, separated by a 28-day period of supplementation: a pre-supplementation trial (seven days after the familiarisation), and a post-supplementation trial (28 days after the pre-supplementation trial). Tests were undertaken towards the end of a regular competition season. Participants were randomly assigned to placebo (PLA, n=11), creatine monohydrate only (Cr, n=10), beta-alanine only (BA, n=11), or beta-alanine plus creatine monohydrate (BACr, n=12) groups.

The familiarisation trial involved anthropometric measurements, including body height (cm, Harpenden stadiometer, UK), body mass (kg) and body fat (%), Tanita BC 418 MA, Tokyo, Japan). In addition, mid-thigh and calf girths were measured with a non-elastic tape measure (Harpenden, UK)

Table 1. Subjects characteristics (mean \pm SD) in the placebo (PLA), creatine alone (Cr), beta-alanine alone (BA), and combined creatine and beta-alanine (BACr) groups (A. football: American football; Δ BL: delta blood lactate concentration)

| | PLA | Cr | BA | BACr | p |
|---|---|---|---|--|------|
| Sample recruited - | 9 males 2 females | 8 males 2 females | 8 males 3 females | 9 males 3 females | - |
| Sport | soccer (n=4) hockey (n=2) A. football (n=2) rugby (n=1) netball (n=1) squash (n=1) | soccer (n=3) hockey (n=1) A. football (n=1) rugby (n=2) basketball (n=2) badminton (n=1) | soccer (n=4) hockey (n=2) A. football (n=2) rugby (n=1), lacrosse (n=1) tennis (n=1) | soccer (n=5) hockey (n=1) rugby (n=1) netball (n=1) tennis (n=1) basketball (n=2) volleyball (n=1) | - |
| Drop-out (reasons) | n=2 (personal reasons) and n=1 (injury#) | n=1 (injury#) | n=2 (paraesthesia) | n=2 (injury#) | |
| Final sample | 6 males 2 females | 7 males 2 females | 7 males 2 females | 8 males 2 females | |
| Training experience (years) | 5.2 \pm 1.6 | 5.8 \pm 1.9 | 4.4 \pm 2.1 | 5.0 \pm 1.9 | |
| Age (years) | 25.2 \pm 1.6 | 25.6 \pm 2.5 | 24.2 \pm 2.2 | 26.0 \pm 1.4 | .545 |
| Height (cm) | 174.9 \pm 8.9 | 173.8 \pm 9.7 | 176.0 \pm 8.0 | 176.8 \pm 7.9 | .477 |
| Body mass (kg) | Pre 75.6 \pm 8.4 Post 75.0 \pm 9.0 | Pre 74.7 \pm 6.6 Post 77.3 \pm 6.8* | Pre 77.1 \pm 9.0 Post 77.2 \pm 8.8 | Pre 76.4 \pm 9.2 Post 77.3 \pm 8.9 | .032 |
| Calf girth (cm) | Pre 35.3 \pm 2.4 Post 35.1 \pm 1.9 | Pre 37.2 \pm 1.7 Post 38.1 \pm 1.4 | Pre 36.7 \pm 1.5 Post 37.0 \pm 1.5 | Pre 38.8 \pm 2.1 Post 38.8 \pm 2.5 | .345 |
| Thigh girth (cm) | Pre 51.6 \pm 3.8 Post 50.8 \pm 4.3 | Pre 52.2 \pm 4.2 Post 53.4 \pm 4.1 | Pre 54.5 \pm 4.2 Post 54.1 \pm 4.3 | Pre 55.5 \pm 4.0 Post 55.0 \pm 3.9 | .242 |
| Body fat (%) | Pre 15.8 \pm 5.4 Post 15.0 \pm 4.9 | Pre 14.7 \pm 6.1 Post 14.9 \pm 5.9 | Pre 16.9 \pm 6.6 Post 17.2 \pm 7.2 | Pre 14.8 \pm 7.6 Post 15.5 \pm 8.4 | .714 |
| Δ BL (mmol·L⁻¹) | Pre 12.8 \pm 2.4 Post 12.8 \pm 3.6 | Pre 14.1 \pm 3.2 Post 14.2 \pm 3.4 | Pre 14.4 \pm 3.0 Post 15.3 \pm 4.1 | Pre 12.5 \pm 2.9 Post 13.6 \pm 3.4 | .276 |

Note. *: significantly different from pre-supplementation values, $p < .05$; #: injuries were independent of the study.

according to the recommendations from the International Society for the Advancement of Kinanthropometry (ISAK). All these measures were taken by a qualified ISAK technician. Subsequently, subjects familiarised themselves with all procedures. To avoid diet-induced variability in exercise performance, all subjects were instructed to complete a 24-h food diary on the day before the first experimental trial and to replicate the same diet during the day before the second experimental trial. Furthermore, participants were instructed to abstain from strenuous exercise, alcohol and caffeine on the day preceding each experimental trial and to abstain from additional nutritional or performance enhancing supplements during the study.

Dietary supplementation

Participants were required to ingest 15 gelatine capsules a day (five at each main meal) for 28 days. Capsules contained either (a) 11 g·day⁻¹ of rice-flour (Doves Farm Rice Flour, Sainsbury's) in 15 PLA capsules, (b) 6 g·day⁻¹ of BA (MyProtein, UK) in seven BA capsules plus eight PLA capsules, (c) 5 g·day⁻¹ of creatine monohydrate (MyProtein, UK) in eight Cr capsules plus seven PLA capsules, and (d) 6 g·day⁻¹ of BA and 5 g·day⁻¹ of Cr in seven BA capsules plus eight Cr capsules.

Repeated sprint sequence (RSS) protocol

The repeated sprint test protocol used in the present study consisted of ten maximal 6-s sprints on a cycle ergometer (Lode Excalibur Sport, Groningen, The Netherlands), interspersed by 24 s of passive recovery periods, against a load corresponding to 7.5% of participants' body mass (Bishop & Spencer, 2004). Participants were verbally encouraged to produce maximal effort throughout the test. In addition, a criterion score assessment was performed before the main test, to avoid pacing (Bishop & Spencer, 2004).

Outcome measures

The power averaged over the 6-s of each sprint was automatically computed (Lode Ergometry Manager software®, Groningen, The Netherlands). Then, these values were used to calculate peak power (PP, in W), defined as the maximal value over the ten sprints, and mean power (MP, in W), calculated as the average power over the ten sprints. These values were then expressed relative to participants' body mass (W·kg⁻¹). Finally, performance decrement was also computed: %Dec=100-([mean power/peak power] x 100) (Fitzimons, Dawson, Ward, & Wilkinson, 1993).

Heart rate (HR, in beats·min⁻¹) was continuously measured during the test (Polar V800, Kempe, Finland), and averages during each 6-s sprint (HR_{mean}; beats·min⁻¹) and during the last 6-s of recovery following each sprint (HR_{recov}; beats·min⁻¹) were calculated. In addition, immediately after each sprint, subjects' rate of perceived exertion (RPE) was recorded using the Borg scale (Borg, 1982). Finally, approximately 5µL fingertip capillary blood samples were collected from fingertips at rest and within one minute after the completion of sprint 10. They were analysed for blood lactate concentration (LA, Lactate Pro 2, Arkray, Japan), and the difference between rest and post-exercise was calculated (Δ LA, in mmol·L⁻¹).

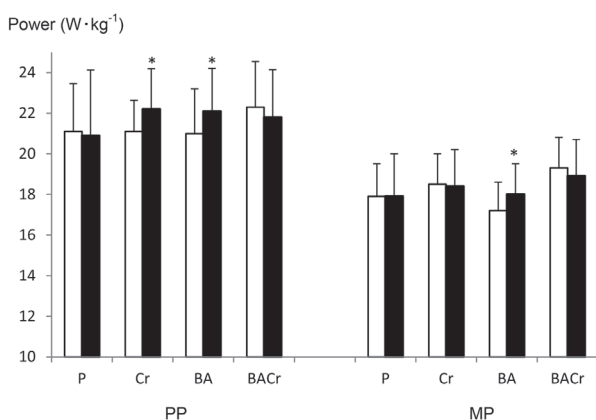
Statistical analyses

All data are reported as mean \pm standard deviation (SD). The Shapiro-Wilks test indicated normal distributions, and consequently a one-way analysis of variance (ANOVA) was used to examine differences between groups in baseline characteristics (age, body height, body mass, and %body fat). In addition, ANOVAs with repeated measures followed by Bonferroni pairwise comparisons were performed to assess the effect of groups, time and sprint number (the latter only for HR and RPE) on outcome variables. The level of significance was set at $p < .05$. Effect sizes were calculated using partial eta squared (η_p^2) and Cohen's d (d) and interpreted according to Cohen (1988).

Results

Performance variables

No significant differences between the groups were shown before the supplementation for any of the variables ($p > .05$).



Note. *: significantly different from pre-supplementation, $p < .05$.

Figure 1. Differences in peak power (PP) and mean power (MP) between the pre- (white) and post- (black) supplementation periods (P: placebo; Cr: creatine; BA: beta-alanine; BACr: creatine and beta-alanine combination).

The statistical analysis showed a significant effect of time ($p = .006$, $\eta_p^2 = 0.179$) and interaction between time and group ($p = .049$, $\eta_p^2 = 0.180$) on PP. Significant increases in PP in the post- compared to the pre-supplementation periods were observed in the Cr (+5.2%, $p = .014$, $d = 0.62$, 95% CI: 0.10 to 1.88 W·kg⁻¹) and BA (+5.3%, $p = .010$, $d = 0.50$, 95% CI: 0.35 to 1.96 W·kg⁻¹) groups only (Figure 1). In contrast, no significant increase in PP was observed in the P and BACr groups.

Despite no significant effect of time ($p = .144$, $\eta_p^2 = 0.053$) or group ($p = .294$, $\eta_p^2 = 0.090$) on MP, a significant interaction between these factors was shown ($p = .046$, $\eta_p^2 = 0.180$). *Post-hoc* comparisons showed a significant increase in MP in the BA group only (+4.7%, $p = .023$, $d = 0.52$, 95% CI: 0.48 to 1.55 W·kg⁻¹, Figure 1). Finally, no significant effect of time ($p = .059$, $\eta_p^2 = 0.095$), group ($p = .310$, $\eta_p^2 = 0.087$), or their interaction ($p = .169$, $\eta_p^2 = 0.120$) was observed on %Dec, with values ranging from 11.9 \pm 3.9% to 15.0 \pm 6.3% in the pre-test, and from 13.6 \pm 6.6 to 18.6 \pm 6.0 in the post-test. Figure 2 shows the profile of power outputs for each group in the pre- and post-tests.

Physiological, anthropometric, and perceptual variables

No significant effect of time ($p = .276$, $\eta_p^2 = 0.033$), group ($p = .357$, $\eta_p^2 = 0.085$), or time*group interaction ($p = .773$, $\eta_p^2 = 0.030$) was observed on Δ LA and calf or thigh girths (Table 1). A significant effect of sprint number only was shown on RPE ($p = .001$, $\eta_p^2 = 0.854$), with RPE significantly increasing with each sprint (RPE ranging from 11.1 \pm 2.9 to 13.2 \pm 2.7 in sprint 1 to 18.2 \pm 1.2 to 19.1 \pm 1.0 in sprint 10, $p = .001$).

A significant effect of sprint number ($p = .001$, $\eta_p^2 = 0.658$) and the interaction between sprint number, group, and time ($p = .023$, $\eta_p^2 = 0.121$) were shown on HR_{mean}. In the PLA, BA, and BACr groups, significant effects of sprint number only were observed ($p = .001$, $\eta_p^2 = 0.648$ to 0.683). However, the Cr group was characterised by significant effects of sprint number ($p = 0.001$, $\eta_p^2 = 0.686$), as well as the interaction between sprint number and time ($p = .001$, $\eta_p^2 = 0.421$). A significant effect of sprint number was shown on HR_{recov} ($p = .001$, $\eta_p^2 = 0.733$). Pairwise comparisons between sprints in each condition for both variables are shown in Figures 3-a to 3-d.

Discussion and conclusions

The main findings of the present study were the significant positive effects of Cr alone on peak power and BA alone on peak and mean power. The greater power outputs observed occurred without any significant differences in physiological parameters measured between the groups. However, we did not observe any significant additional benefits

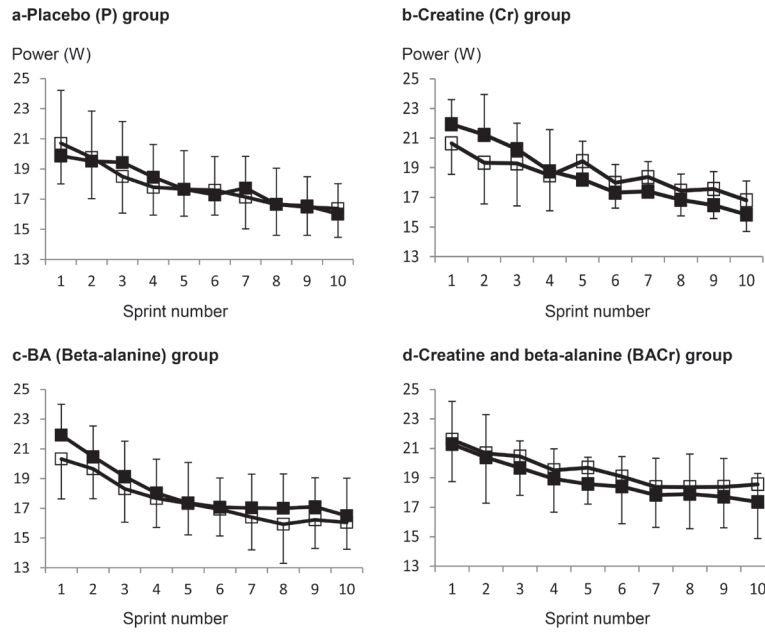
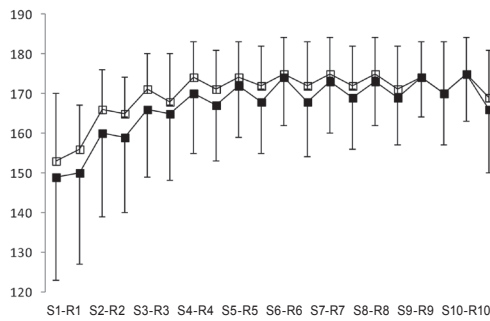


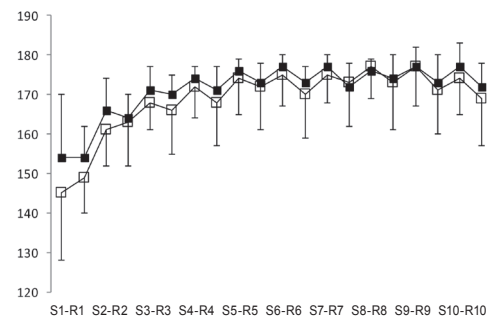
Figure 2. Power output for each sprint of the RSS test in the pre- (empty symbols) and post- (filled symbols) supplementation periods in the P (a), Cr (b), BA (c) and BACr (d) groups.

a-P
HR (beats·min⁻¹)



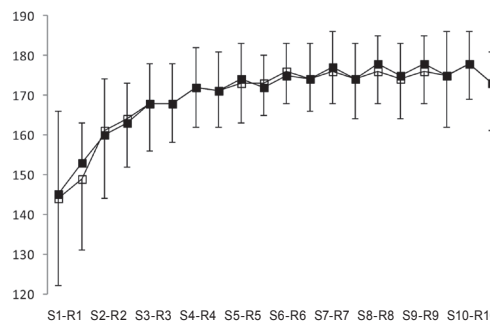
HRmean: significant differences between S1 and S3 to S8, between S2 and S3 to S7, and between S3 and S6, p<.05.
HRrecov: significant differences between S1 to S2 and all the others; between S3 and all but S10; between S4 and all but S9 and S10; and between S8 to S9 and S10, p<.05.

b-Cr
HR (beats·min⁻¹)



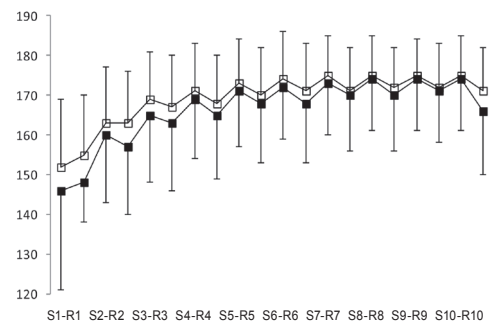
HRmean: Pre-supplementation: significant differences between S1 and S5 to S6, p<.05. Post-supplementation: significant differences between S1 and S3 to S9, and between S3 and S5 and S7 to S8, p<.05.
HRrecov: significant differences between S1 to S2 and all the others; between S3 and all but S10; between S4 and all but S9 and S10; and between S8 to S9 and S10, p<.05.

c-BA
HR (beats·min⁻¹)



HRmean: significant differences between S1 and S4 to S7; between S5 and S6, p<.05.
HRrecov: significant differences between S1 to S2 and all the others; between S3 and all but S10; between S4 and all but S9 and S10; and between S8 to S9 and S10, p<.05.

d-BACr
HR (beats·min⁻¹)



HRmean: significant differences between S1 and all the others, p<.05.
HRrecov: significant differences between S1 to S2 and all the others; between S3 and all but S10; between S4 and all but S9 and S10; and between S8 to S9 and S10, p<.05.

Figure 3. Heart rate during (HR_{mean}) and after (HR_{recov}) each sprint of the RSS test in the pre- (empty symbols) and post- (full symbols) supplementation periods. S1 to S10: sprints 1 to 10; R1 to R10: recovery periods after sprints 1 to 10.

of the combination of Cr and BA, compared to their isolated use, on repeated sprint performance in team and racket sport players.

Significant increases in PP were observed in the post- compared to the pre-supplementation test in the Cr (+5.2%) and BA (+5.3%) groups only, both accompanied by moderate effect sizes. These results are similar to those of Yquel et al. (2002), showing a 5% increase in peak-power output in bouts 3-7 of a 7-bout intermittent-sprint protocol (8-16-s sprints) following a 6-day Cr loading protocol (20 g·day⁻¹), compared to a control condition in healthy men. In addition, van Loon et al. (2003) used a longer supplementation period (five days loading at 20 g·day⁻¹ and six weeks at 2 g·day⁻¹) and observed a significant increase in PP in seven out of the 12 sprints of a twelve 12-s intermittent-sprint protocol. These increases in PP following Cr supplementation are likely to be due to an increase in muscle creatine stores, as it has been shown that resting muscle creatine stores can be increased by up to 20% via exogenous supplementation (Allen, et al., 2008). Intramuscular Cr stores have been shown to be reduced by 57% and 83%, respectively, after the first and the last sprint of a 10x6 second (30-s recovery) repeated-sprint protocol (Gaitanos, et al., 1993). Unfortunately, no measure of intramuscular Cr was obtained in the present study; however, body mass was significantly increased and there was a trend for greater thigh girth measures in the post-compared to the pre-supplementation in the Cr group only. Increased body mass after Cr supplementation protocols is commonly observed in literature and usually accounted for by a rise in fat free mass (e.g., van Loon, et al., 2003). The effects of BA on PP during RSS show more contrasting results in literature, with no significant effect observed during short intermittent sprint protocols, eg. 5x5-s and 6x20-m running sprints (Ducker, et al., 2013; Sweeney, et al., 2010), but significant improvements in PP were observed during longer SIT protocols (4x30-s upper body Wingate tests each separated by 3-min rest periods; Tobias, et al., 2013). One explanation for the significant effect of BA on PP in the present study could be that our sprint duration and number of repetitions were slightly greater than the short sprints described above. Furthermore, it has been suggested that, in addition to its buffering benefits, BA increases calcium sensitivity in contractile fibres (Hobson, et al., 2012), which could in turn result in a greater PP. However, these mechanisms have not been fully determined, and hence further studies are necessary to investigate the effects of BA on RSS.

In the present study, MP was significantly improved in the BA group only (+4.7%), with a moderate effect size. In addition, no significant difference in %Dec was observed in any group. In the context of team or racket sports, the greater PP

and MP in the BA group in the post- compared to the pre-supplementation suggests that this type of supplementation is better than Cr (increase in PP but no change in MP or %Dec) or BACr (no variation in PP, MP, or %Dec) because it would not only increase the quality of sprints at the start of a match, but it would also maintain performance throughout the match. This can be illustrated on the power profiles shown in Figure 2, where the only condition where the post-supplementation powers are over the pre-supplementation powers in the second part of the test is the BA condition. However, the application of our findings to team and racket sports is somewhat limited by the fact that we used a cycling repeated sprint protocol. Literature presents contrasting results about the effects of BA on repeated sprint performance. For instance, a recent study by Tobias et al. (2013) found a significant effect of BA on the mean power in the 2nd and 3rd bouts of a 4x30-s upper body Wingate tests each separated by 3-min rest periods in competitive judo and jiu-jitsu athletes. In contrast, other studies using shorter sprint times found similar results to the present study, with no significant effect of BA supplementation on MP or %Dec (Ducker, et al., 2013; Sweeney, et al., 2010). In addition, results from a meta-analysis suggested that BA does not affect exercise performance in exercises lasting less than 60-s (Hobson, et al., 2012). The benefits of BA supplementation on repeated sprint performance are classically attributed to the buffering of H⁺ ions during the recovery between bouts by carnosine inside the muscle fibre. Indeed, Harris et al. (2006) reported a 64% increase in intramuscular carnosine levels following a 4-week supplementation period with 6.4 g·d⁻¹ of BA. Therefore, sprint duration of 6-s in the present study might not have been sufficient to stimulate the anaerobic glycolysis energy system and produce enough lactate and H⁺ ions, and our recovery might not have been long enough to allow enough buffering to take place. Within this context, in their review of high-intensity interval training, Buchheit and Laursen (2013) highlighted that blood lactate concentration during RSS protocols similar to the present study were lower than during protocols including longer sprints, such as SIT (6-18 mmol·L⁻¹ vs. 16-22 mmol·L⁻¹). The delta blood lactate values obtained in the present study are in accordance with this hypothesis, with no significant difference between the groups, and values from 12.8 to 15.3 obtained amongst conditions (resting blood lactate lower than 2.0 mmol·L⁻¹). However, there was a trend for an increase in blood lactate between the pre- and the post-supplementation periods in the BA and BACr groups only.

Interestingly, despite the benefits of both Cr and BA on PP, and of BA on MP, the combination of these ergogenic aids not only did not result in any significant improvement in PP and MP, but proved

to be worse than each substance implemented isolated. To our knowledge, our study is the first to investigate the potential benefits of this combination on RSS performance. Our results are in accordance with those of studies focusing on longer sprints, either continuous (mechanical work during a sprint at 110% of maximal power; Hill, et al., 2005), or intermittent (two or three repeated 30-s Wingates; Kresta, et al., 2014; Okudan, et al., 2015), showing significant benefits of Cr or BA, but no additional positive effects of the combination of these supplements. However, comparisons with literature are difficult due to the different doses and administration protocols (loading, maintenance periods). There is no physiological reason why Cr and BA would be counteractive to each other (Harris, et al., 2006). Therefore, the lack of positive effect of the BACr combination on performance variables in the present study could be due to the inter-individual variability in response to the BA and Cr supplementation, with maybe more participants with a low response to these supplements in the BACr group, compared to the Cr and BA groups. For example, around 20 to 30% of individuals who undergo Cr supplementation do not increase their muscle Cr stores (Greenhaff, 1997). Exercise and recovery duration may be another potential reason for the lack of significant differences in the BACr group. Zoeller et al. (2007) showed that BACr significantly improved five out of eight indices of cardio-respiratory endurance during an incremental test, including oxygen consumption and power output at the ventilatory and lactate thresholds, while the sole use of Cr or BA was less beneficial. In RSS, the aerobic system is used to replenish PCr stores and metabolise lactate during the recovery between sprints. Therefore, we could hypothesize a greater oxidative capacity after the BACr supplementation that could result in improved RSS performance, as

RSS performance has been correlated to aerobic capacity (Girard, et al., 2011). Therefore, recovery periods (24-s) between the sprints might not have been long enough for a maximal involvement of the aerobic system to restore PCr stores or clear lactate. In favour of this hypothesis, a similar study by Kamber et al. (1999), which involved longer recovery periods than 24-s (i.e. 30-s) found 7% improvement in performance in the late phases of repeated sprints (10x6-s) following Cr supplementation. Finally, we cannot exclude a potential effect of fatigue or overtraining in some of our participants, since testing was undertaken towards the end of the season.

The main limitations of the present study were a rather small sample size and relatively high drop-out rate; however these numbers are expected in 4-week long studies and our sample was similar or greater than in other studies in this area (e.g., Kresta, et al., 2014). In addition, measuring the amount of creatine or carnosine in muscles would have helped interpret our findings. Finally, the extrapolation of our findings to team and racket sports is limited by the fact that our protocol was performed on a bicycle ergometer.

In conclusion, our main results did not show any significant additional benefits of the combination of Cr and BA, compared to their isolated use on RSS. However, the positive effects of Cr alone on PP as well as BA alone on PP and MP highlight that these substances are ergogenic for these types of sports when consumed in isolation. While the combination of Cr and BA might be recommended for longer repeated sprints or endurance events, it may not be for short repeated sprints characteristic of team and racket sports. Further studies should be undertaken to understand the mechanisms involved in the effects of BA, particularly on performance during RSS.

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Submitted: October 2, 2018

Accepted: October 10, 2018

Published Online First: May 15, 2020

Corresponding author:

Anne Delextrat, Ph.D.

Movement Science Group, Oxford Brookes University

Gypsy lane, Oxford OX3 0BP, United Kingdom

Email: adelextrat@brookes.ac.uk

Tel: +44 (0) 1865 48 3610

Fax: +44 (0) 1865 48 3242

Acknowledgment, authorships and declarations.

The authors would like to thank all participants for their time. This research was not funded. There were no conflicts of interest.