No differences between beetroot juice and placebo on competitive 5-km running performance: A double-blind, placebo-controlled trial

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Running head: Beetroot juice and competitive running performance

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Abstract

We examined the effect of beetroot juice on endurance running performance in "real-world" competitive settings. One-hundred recreational runners (54% male; mean ± standard deviation, age = 33.3 ± 12.3 years, training history = 11.9 ± 8.1 years, hours per week training = 5.9 ± 3.5) completed a quasi-randomised, double-blind, placebo-controlled study of 5-km competitive time-trials. Participants performed four trials separated by one week in the order of pre-baseline, two experimental, and one post-baseline. Experimental trials consisted of the administration of 70-mL nitrate rich beetroot juice (containing ~4.1 mmol of nitrate, Beet It Sport®) or nitrate depleted placebo (containing ~0.04 mmol of nitrate, Beet It Sport®) 2.5 hours prior to time-trials. Time to complete 5-km was recorded for each trial. No differences were shown between pre- and post-baseline ($P = 0.128$, CV = 2.66%). The average of these two trials is therefore used as baseline. Compared to baseline, participants ran faster with beetroot juice (mean differences = 22.2 ± 5.0 s, $P < 0.001$, $d = 0.08$) and placebo (22.9 ± 4.5 s, $P < 0.001$, $d = 0.09$). No differences in times were shown between beetroot juice or placebo (0.8 ± 5.7 s, $P < 0.875$, $d = 0.00$). These results indicate that an acute dose of beetroot juice does not improve competitive 5-km time-trial performance in recreational runners compared to placebo.

Keywords: dietary nitrate, ecological validity, ergogenic aids, nutrition, sport supplements

Introduction

Dietary nitrate supplementation increases plasma nitrate and nitrite via nitric oxide synthase independent pathway (Kapil et al., 2010) and has been shown to reduce blood pressure (Vanhatalo et al., 2010), adenosine triphosphate utilisation, phosphocreatine degradation (Bailey, Fulford, et al., 2010), the oxygen cost of submaximal exercise (Muggeridge et al., 2013; Wylie et al., 2016) and improve sport performance (Hoon et al., 2013; McMahon et al., 2017). In the last decade, there has been an exponential increase in research investigating the ergogenic effects of dietary nitrate rich products, such as beetroot juice (Hoon et al., 2013; Jones, 2014; McMahon et al., 2017).

Dietary nitrate supplementation is a popular ergogenic aid amongst athletes of all abilities (Garthe & Maughan, 2018; Maughan et al., 2018). While a growing body of research has investigated the effects of dietary nitrate in elite athletes (Cermak, Gibala, et al., 2012; Cermak, Res, et al., 2012; Peeling et al., 2015), most research has sampled recreational cohorts (Hoon et al., 2013; McMahon et al., 2017). Bailey, Winyard, et al. (2010) examined the effects of dietary nitrate on time-to-exhaustion during graded step exercise in recreationally active participants (N = 7) and reported improvements of 16% compared to placebo. Similarly, Vanhatalo et al. (2010) reported that both acute (one day) and chronic (15 days) 0.5-L dietary nitrate supplementation improved steady-state $\dot{V}O_2$ during moderate-intensity exercise by ~4% in healthy participants (N = 8) and Jodra et al. (2020) showed that consumption of a 70-mL beetroot juice shot improved peak power-output during a Wingate test by 4% in recreationally trained participants (N = 15).

While data suggests dietary nitrate can improve sport performance (Hoon et al., 2013; Jones, 2014; McMahon et al., 2017), there are three limitations that characterise the literature. First, studies often assess performance in tightly controlled laboratories (Hoon et al., 2013; McMahon et al., 2017) and it is unknown whether the effects are similar in real-world competitive events. Second, testing often takes place in isolation with participants performing alone. It is well known that improvements in performance are shown during competition than exercising alone (Cooke et al., 2011; Corbett et al., 2012; Williams et al., 2015). It is therefore understandable to suggest that the beneficial effects of dietary nitrate and competition may not be additive and less marked during competition. Third, although studies may be sufficiently powered, two meta-analyses (Hoon et al., 2013; McMahon et al., 2017) report that studies investigating the effectiveness of dietary nitrate on sport performance often use small sample sizes (mean N = 11), which limit the detection of meaningful changes on performance (Burke & Peeling, 2018).

Given the above, and to progress knowledge and understanding of the effectiveness of dietary nitrate on sport performance, we aimed to determine the effect of dietary nitrate in the form of beetroot juice on sport performance during a competitive time-trial using a sufficiently large sample. We used parkrun® as our time-trial event, which has shown to be a highly reliable measure of 5-km running performance (CV = 0.95%; Hurst & Board, 2017). Since 2004, parkrun has established weekly, free, 5-km running events that take place in more than 650 locations globally, with some events hosting over 1000 runners (parkrun, 2020). We used a double-blind, quasi-randomised, placebo-controlled trial to investigate the effect of an acute dose of beetroot juice on time to complete a 5-km parkrun time-trial. We hypothesised that beetroot juice would improve time to complete 5-km compared to baseline and placebo.

Methods

The reporting of the current study followed the Proper Reporting of Evidence in Sport & Exercise Nutrition Trials (PRESENT) 2020 checklist (Betts et al., 2020).

Participants

One-hundred recreational runners were recruited to the study. Of these participants, 25 did not complete all trials and five reported injuries affecting their performance. These were removed leaving a final sample size of 70. Demographics for participants are shown in Table 1. A minimum sample size of 66 was calculated to detect a medium effect of beetroot juice on time to complete a 5-km time-trial. This sample was determined by power analysis using the G*Power v3.1 software (Faul et al., 2009), using a repeated measures ANOVA design, in which significance was set at 0.05, power (1-beta) at 95%, and given that effect sizes greater 0.2 are considered potentially beneficial for sport performance (Hopkins et al., 1999), the effect size (F) at 0.2.

Inclusion criteria stipulated that participants had to be 18 years or over, passed a health questionnaire and have no indication of a physical injury. In addition, Hurst and Board (2017) reported that participants with greater familiarity of the parkrun course are more likely to improve test-retest reliability and reduce the coefficient variation (CV) of the performance measure. Thus, inclusion criteria stipulated that participants had completed two or more parkruns in the last four weeks and five or more in the preceding six months. The average number of parkruns participants performed at the time of recruitment was 24 ± 21.

Design

We used a within-participant, quasi-randomised, double-blind, placebo-controlled trial to determine the effects of an acute dose of beetroot juice on competitive 5-km running performance. Participants performed four trials separated by one week in the order of pre-baseline, two experimental, and one post-baseline. In experimental trials, participants were randomly allocated (1:1 ratio, no blocking or stratification) to receive beetroot juice or placebo using a computer-generator programme (www.randomization.org).

Supplementation

Participants consumed concentrated nitrate rich beetroot juice (containing ~4.1 mmol of nitrate; Beet it, James White Drinks Ltd., Ipswich, UK) and nitrate depleted beetroot juice (organic beetroot juice containing ~0.04 mmol of nitrate; Beet it, James White Drinks Ltd., Ipswich, UK). Pharmacokinetic data report that plasma nitrate peaks between 2.5 – 3 hours after ingestion of a single dose of beetroot juice (Webb et al., 2008), thus on the day of experimental trials, participants were instructed to consume 70-mL of the supplement 2.5 hours before the beginning of the trial. Both supplements were indistinguishable in taste and smell. Pilot testing with six participants not involved in the main study, were unable to identify which supplement had been ingested. The packaging of both supplements were identical in appearance, which were marked by a researcher with a unique code (i.e. “X” or “Y’) for random assignment. One researcher, who was not involved with any experimental testing, knew which codes corresponded to each supplement. To ensure that the placebo blind had been effective, a manipulation check was conducted after each experimental time-trial. Participants were asked to state what supplement they had received by selecting one of three options: 1) beetroot juice; 2) placebo and; 3) don’t know. Participants also indicated what time they had taken the shot, if any habitual practices in training and diet had changed leading up to the trial and if any other factors (e.g. motivation to perform the trial as fast as possible, weather conditions and injuries) affected their performance on the day of the trial.

Procedure

Ethical approval was granted by the lead author’s Institutional Ethics Committee (ref: 14/SAS/189) and parkrun’s Ethics Committee in accordance with the Declaration of Helsinki. Participants were recruited to the study in person and informed about the study’s aim, that participation was voluntary, and that all data collected would be used for research purposes only. After reading the information sheet and completing a health questionnaire, written informed consent was obtained.

All trials were performed on a Saturday morning at 09:00 at the same location in Kent, United Kingdom between April and May 2015. Ambient conditions were recorded using publicly available data (https://www.wunderground.com/) collected by The Weather Company (IBM, Atlanta, Georgia, USA). Minimal differences were reported for all time-trials (temperature = 11.2 ± 1.8°C; humidity = 66 ± 4%; and windspeed = 14.6 ± 2 km/hr). Participants were instructed to keep exercise and nutritional habits the same, refrain from alcohol 24 hours preceding the trial, high intensity exercise 48-hours prior to the trials and requested not to consume other sport supplements not associated with the study. Participants were instructed to run the 5-km as fast as possible. Trials were performed alongside other runners not involved with the trial. Volunteer parkrun officials recorded completion times with data extracted from the official website at a later date (parkrun, 2020). Upon completion, participants reported to the research team who provided instructions for the next trial.

Data analysis

Time to complete 5-km for baseline trials were inputted into an online reliability spreadsheet to estimate reliability of pre- and post-baseline trials. Data was log transformed to reduce nonuniform errors and Pearson correlation (*r*), the intraclass correlation (ICC) and CV provided estimates of reliability. The *r* coefficient was interpreted as trivial (<0.1), small (0.3), moderate (0.5), large (0.7), nearly perfect (0.9) and perfect (1.0; Hopkins, 2015). The ICC was interpreted as low (0.20), moderate (0.50), high (0.75), very high (0.90) and extremely high (0.99; Hopkins, 2015). A paired samples *t*-test was conducted to determine systematic differences in performance between baseline trials.

Data was analysed using SPSS version 24.0 (IBM, Armonk, NY) and tested for homogeneity of variance, normal distribution and outliers. Ratings of supplement assignment (correct, incorrect) were analysed using Chi-square (χ^2). Cramer's V was used as the effect size and interpreted as 0.10, 0.30 and 0.50, for a small, medium and large effect, respectively (Cohen, 2013). Repeated measures analysis of variance (ANOVA) was conducted to analyse effects of time between conditions. Greenhouse-Geisser epsilon was reported when sphericity was violated. Partial eta-squared (η^2) is reported as the effect size, with values of 0.02, 0.13 and 0.26 indicating small, medium and large effects respectively (Cohen, 1992). Post-hoc Least Significant Difference (LSD) tests were used to examine differences between conditions and Cohen's *d* (*d*) was calculated with values 0.2, 0.5 and 0.8 indicating small, medium and large effects, respectively (Cohen, 1992). Data is reported as means ± standard error of the mean (SEM) and 95% confidence intervals. Statistical significance was set at P <0.05.

Results

Preliminary analyses

Times were similar between pre- and post-baseline (mean differences = 16.15 ± 1.47 s, 95% CI = -4.80 to 37.10 s, *P* = 0.128, *r* = 0.95, ICC = 0.95, CV = 2.66%). The average of these two time-trials was thus used to measure baseline.

Main analyses

Results of χ^2 tests indicated that participants did not accurately guess whether they were given beetroot juice or placebo ($\chi^2 = 49.352$, $P = 0.457$, Cramer's $V = 0.09$). All participants reported to consume the supplement 2.5 hours before the start of the time-trial for each condition and none reported differences in training and nutritional routines leading up to the trials or factors affecting their performance (i.e. injuries, motivation and weather).

Mean times for each condition are shown in figure 1. Repeated measures ANOVA revealed a significant effect for 5-km time between each condition ($F_{2, 138} = 13.075$, $P < 0.001$, $\eta^2 = 0.159$). Compared to baseline, participants ran faster in the beetroot (mean differences = 22.2 ± 5.0 s, 95% CI = 12.2 to 32.1 s $P < 0.001$, $d = 0.08$) and placebo (22.9 ± 4.5 s, 95% CI = 13.9 to 32.0 s, $P < 0.001$, $d = 0.09$) conditions. No differences in times were reported between beetroot and placebo (0.8 ± 5.7 s, 95% CI = -10.6 to 12.1 s*,* $P = 0.875$, $d = 0.00$).

Discussion

This study was a first to use a double-blind, quasi-randomised, placebo-controlled trial to determine the effect of an acute dose of beetroot juice on competitive 5-km running performance in recreational runners. Our results indicate that compared to baseline, beetroot juice improves performance by on average 22.2 seconds (1.4%). However, when compared to a placebo, performance did not change, with mean differences reported at 0.8 seconds (0.05%). Collectively, results suggest that an acute does of beetroot juice does not improve 5-km performance in recreational runners.

While meta-analyses report beneficial effects of beetroot juice on endurance performance (Hoon et al., 2013; McMahon et al., 2017), we found that beetroot juice does not improve time to complete a

5-km time-trial. These results are similar to Cermak, Res, et al. (2012) and de Castro et al. (2019) who reported that compared to placebo, beetroot juice supplementation does not improve 1-hour cycling time-trial and time to complete 10-km running trial performance, respectively. More recent research (Jodra et al., 2020; Jonvik et al., 2018; Shannon et al., 2017) has reported that beetroot juice is more likely to affect shorter (e.g. 1500-m running) than longer distance (e.g. 10,000-m running) events. Shannon et al. (2017) suggest that dietary nitrate supplementation increases the recruitment of type II muscle fibres and augments blood flow and oxygen delivery. The increase in local blood flow is argued to decrease metabolic perturbations such as PCr degradation and adenosine diphosphate (ADP) accumulation (Vanhatalo et al., 2011), increase muscle force production and ultimately performance (Coggan et al., 2015). Thus, these effects are less likely to impact endurance performance. Given the results of our study, beetroot juice may have little effect on 5-km running time-trial performance.

The null effects could also be explained by our main outcome variable. To help maximise the validity of our findings, we used an outdoor competitive 5-km time-trial. The physiological effects associated with beetroot juice may not influence performance as much during competitive time-trials than other factors (e.g. social comparisons, rewards for success and anxiety). While a 5-km parkrun may not produce the same psychophysiological response as the Olympics and World Championships, the results of our study are an important first step in identifying whether an acute dose of beetroot juice improves endurance performance in an ecological valid setting. Given that recreational runners arguably account for a substantial proportion of the consumer group for nutritional sport supplements (Maughan et al., 2018), our results highlight that the physiological effects of beetroot juice are unlikely to improve performance for this population. Instead, recreational runners should practice other methods that are more likely to benefit their performance in competitive settings (e.g. an improved training programme, nutritional strategy or psychological profile).

It is important to consider the reliability of the performance measure when interpreting results. We reported improvements compared to baseline of 1.4% for both the beetroot juice and placebo condition. However, the CV of our measure was 2.66%. It is therefore likely that changes are attributable to systematic and random error. Similarly, the CV of our study is greater than previous research using a similar performance measure (CV = 0.95%; Hurst & Board, 2017). Reasons for the larger variance could be related to the time in-between baseline trials. Hurst and Board (2017) measured 5-km performance twice, separated by 1-week, whereas we separated baseline trials by 3-weeks. Although no differences were shown between baseline trials, it could be speculated that the greater time in-between trials increased the variance in our performance measure. This highlights the importance of measuring a further baseline time-trial after experimental trials to help identify systematic and random error of performance.

While our performance measure is not as reliable as previous research (Hurst & Board, 2017), the performance measure still holds very good reliability (see Currell & Jeukendrup, 2008). Therefore, the results of our study are supported with high reliability and validity, and a large sample size. Generally, randomised controlled trials in sport and exercise employ small sample sizes and use outcome measurements in tightly controlled laboratories (Burke & Peeling, 2018). This approach can cause difficulties for researchers detecting meaningful changes in performance and translating the findings to applied practice. While challenges exist in recruiting adequate sample sizes and designing studies that are both reliable and valid, the results of this study highlight the opportunity for researchers to analyse the effects of interventions using a reliable and valid measure of running performance with a large sample. By using parkrun as our outcome measure, and recruiting a large sample, this study offers a clearer estimate of the true magnitude of changes in 5-km running performance after administration of an acute dose of beetroot juice.

Limitations and future research

While the study has a number of strengths relating to the study design, sample size and outcome measure, there were limitations. First, we measured the effect of a single acute dose of beetroot juice (70-mL). There is evidence to suggest that chronic supplementation of beetroot juice may be more beneficial for improving sport performance than acute supplementation (Jones, 2014; McMahon et al., 2017). Future research should aim to determine the effect of chronic beetroot juice supplementation on competitive 5-km running performance. Second, we did not control the content of nitrate rich foods (e.g. beetroot, lettuce and spinach) in participants' diet. Those with a higher nitrate rich diet may show reduced effects with beetroot juice supplementation than those with a low nitrate rich diet (Jones, 2014; Jonvik et al., 2017). Prospective research should consider controlling for the impact of the consumption of nitrate rich diets in their results. Third, while we recruited a large sample size that were regular 5-km runners, they were not elite athletes. It is argued that the benefits of beetroot juice supplementation are more likely to be shown for highly-trained competitive athletes than recreational athletes due to the consequence of years of training adaptations and genetic factors (Burke & Peeling, 2018). Future research should aim to sample more highly trained athletes to further elucidate the effects of beetroot juice on competitive running performance. Fourth, given that our outcome measure does not mimic the atmosphere, pressure and demands that may be experienced during competitive events (e.g. national and international championships), and that athletes did not adjust their training to "peak" for each trial, the "competitive" element of our study is limited. It would be worthwhile to understand the effects of an acute dose of beetroot juice on running performance during more competitive events.

Conclusion

In conclusion, our results indicate that there is no difference in competitive 5-km time-trial performance when participants ingest an acute dose of beetroot juice or an equivalent placebo. This suggests that beetroot juice may not exert an ergogenic effect on 5-km running performance for recreational runners. The results of this study are supported with high reliability and validity using a large sample size. Future research studies should consider using other parkrun events to investigate the effectiveness of other sport interventions.

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Declarations

Authors received no external funding for this research and declare no conflicts of interest.

Authorships

The study was designed by PH and SS; data were collected by PH and SS; data were analysed by PH; data interpretation and manuscript preparation were undertaken by PH, SS and DC. All authors approved the final version of the paper.

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Tables

Table 1 Demographics of participants separated by gender

	Male	Female	Overall
N	38	32	70
Age (years)	34.4 ± 11.6	32.1 ± 12.9	33.3 ± 12.3
Training history (years)	11.8 ± 7.0	11.9 ± 9.5	11.9 ± 8.1
Hours per week training	6.3 ± 3.9	5.5 ± 3.1	5.9 ± 3.5
Number of parkruns	21 ± 18	28 ± 24	24 ± 21
Personal best (minutes: seconds)	23:02 ± 4:42	29:05 ± 3:51	25:48 ± 5:16

Note: data are mean ± standard deviation

Figure

Figure 1. Mean time to complete 5-km time-trials for each condition. Data are means ± SEM. * = P <0.001 vs. beetroot and placebo.