

## Research Space

Journal article

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

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1 **No differences between beetroot juice and placebo on competitive 5-km running performance: A**

2 **double-blind, placebo-controlled trial**

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9 **Running head:** Beetroot juice and competitive running performance

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## 20Abstract

21We examined the effect of beetroot juice on endurance running performance in “real-world”  
22competitive settings. One-hundred recreational runners (54% male; mean  $\pm$  standard deviation, age =  
2333.3  $\pm$  12.3 years, training history = 11.9  $\pm$  8.1 years, hours per week training = 5.9  $\pm$  3.5) completed a  
24quasi-randomised, double-blind, placebo-controlled study of 5-km competitive time-trials.  
25Participants performed four trials separated by one week in the order of pre-baseline, two  
26experimental, and one post-baseline. Experimental trials consisted of the administration of 70-mL  
27nitrate rich beetroot juice (containing  $\sim$ 4.1 mmol of nitrate, Beet It Sport<sup>®</sup>) or nitrate depleted  
28placebo (containing  $\sim$ 0.04 mmol of nitrate, Beet It Sport<sup>®</sup>) 2.5 hours prior to time-trials. Time to  
29complete 5-km was recorded for each trial. No differences were shown between pre- and post-  
30baseline ( $P = 0.128$ , CV = 2.66%). The average of these two trials is therefore used as baseline.  
31Compared to baseline, participants ran faster with beetroot juice (mean differences = 22.2  $\pm$  5.0 s,  $P <$   
320.001,  $d = 0.08$ ) and placebo (22.9  $\pm$  4.5 s,  $P < 0.001$ ,  $d = 0.09$ ). No differences in times were shown  
33between beetroot juice or placebo (0.8  $\pm$  5.7 s,  $P < 0.875$ ,  $d = 0.00$ ). These results indicate that an  
34acute dose of beetroot juice does not improve competitive 5-km time-trial performance in  
35recreational runners compared to placebo.

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



### 38Introduction

39Dietary nitrate supplementation increases plasma nitrate and nitrite via nitric oxide synthase  
40independent pathway (Kapil et al., 2010) and has been shown to reduce blood pressure (Vanhatalo et  
41al., 2010), adenosine triphosphate utilisation, phosphocreatine degradation (Bailey, Fulford, et al.,  
422010), the oxygen cost of submaximal exercise (Muggeridge et al., 2013; Wylie et al., 2016) and  
43improve sport performance (Hoon et al., 2013; McMahon et al., 2017). In the last decade, there has  
44been an exponential increase in research investigating the ergogenic effects of dietary nitrate rich  
45products, such as beetroot juice (Hoon et al., 2013; Jones, 2014; McMahon et al., 2017).

46Dietary nitrate supplementation is a popular ergogenic aid amongst athletes of all abilities (Garthe &  
47Maughan, 2018; Maughan et al., 2018). While a growing body of research has investigated the effects  
48of dietary nitrate in elite athletes (Cermak, Gibala, et al., 2012; Cermak, Res, et al., 2012; Peeling et  
49al., 2015), most research has sampled recreational cohorts (Hoon et al., 2013; McMahon et al., 2017).  
50Bailey, Winyard, et al. (2010) examined the effects of dietary nitrate on time-to-exhaustion during  
51graded step exercise in recreationally active participants (N = 7) and reported improvements of 16%  
52compared to placebo. Similarly, Vanhatalo et al. (2010) reported that both acute (one day) and  
53chronic (15 days) 0.5-L dietary nitrate supplementation improved steady-state  $\dot{V}O_2$  during moderate-  
54intensity exercise by ~4% in healthy participants (N = 8) and Jodra et al. (2020) showed that  
55consumption of a 70-mL beetroot juice shot improved peak power-output during a Wingate test by  
564% in recreationally trained participants (N = 15).

57While data suggests dietary nitrate can improve sport performance (Hoon et al., 2013; Jones, 2014;  
58McMahon et al., 2017), there are three limitations that characterise the literature. First, studies often  
59assess performance in tightly controlled laboratories (Hoon et al., 2013; McMahon et al., 2017) and it  
60is unknown whether the effects are similar in real-world competitive events. Second, testing often  
61takes place in isolation with participants performing alone. It is well known that improvements in  
62performance are shown during competition than exercising alone (Cooke et al., 2011; Corbett et al.,

632012; Williams et al., 2015). It is therefore understandable to suggest that the beneficial effects of  
64dietary nitrate and competition may not be additive and less marked during competition. Third,  
65although studies may be sufficiently powered, two meta-analyses (Hoon et al., 2013; McMahon et al.,  
662017) report that studies investigating the effectiveness of dietary nitrate on sport performance often  
67use small sample sizes (mean N = 11), which limit the detection of meaningful changes on  
68performance (Burke & Peeling, 2018).

69Given the above, and to progress knowledge and understanding of the effectiveness of dietary nitrate  
70on sport performance, we aimed to determine the effect of dietary nitrate in the form of beetroot  
71juice on sport performance during a competitive time-trial using a sufficiently large sample. We used  
72parkrun® as our time-trial event, which has shown to be a highly reliable measure of 5-km running  
73performance (CV = 0.95%; Hurst & Board, 2017). Since 2004, parkrun has established weekly, free, 5-  
74km running events that take place in more than 650 locations globally, with some events hosting over  
751000 runners (parkrun, 2020). We used a double-blind, quasi-randomised, placebo-controlled trial to  
76investigate the effect of an acute dose of beetroot juice on time to complete a 5-km parkrun time-  
77trial. We hypothesised that beetroot juice would improve time to complete 5-km compared to  
78baseline and placebo.

## 79Methods

80The reporting of the current study followed the Proper Reporting of Evidence in Sport & Exercise  
81Nutrition Trials (PRESENT) 2020 checklist (Betts et al., 2020).

### 82Participants

83One-hundred recreational runners were recruited to the study. Of these participants, 25 did not  
84complete all trials and five reported injuries affecting their performance. These were removed leaving  
85a final sample size of 70. Demographics for participants are shown in Table 1. A minimum sample size  
86of 66 was calculated to detect a medium effect of beetroot juice on time to complete a 5-km time-  
87trial. This sample was determined by power analysis using the G\*Power v3.1 software (Faul et al.,

882009), using a repeated measures ANOVA design, in which significance was set at 0.05, power (1-  
89beta) at 95%, and given that effect sizes greater 0.2 are considered potentially beneficial for sport  
90performance (Hopkins et al., 1999), the effect size (F) at 0.2.

91Inclusion criteria stipulated that participants had to be 18 years or over, passed a health questionnaire  
92and have no indication of a physical injury. In addition, Hurst and Board (2017) reported that  
93participants with greater familiarity of the parkrun course are more likely to improve test-retest  
94reliability and reduce the coefficient variation (CV) of the performance measure. Thus, inclusion  
95criteria stipulated that participants had completed two or more parkruns in the last four weeks and  
96five or more in the preceding six months. The average number of parkruns participants performed at  
97the time of recruitment was  $24 \pm 21$ .

#### 98*Design*

99We used a within-participant, quasi-randomised, double-blind, placebo-controlled trial to determine  
100the effects of an acute dose of beetroot juice on competitive 5-km running performance. Participants  
101performed four trials separated by one week in the order of pre-baseline, two experimental, and one  
102post-baseline. In experimental trials, participants were randomly allocated (1:1 ratio, no blocking or  
103stratification) to receive beetroot juice or placebo using a computer-generator programme  
104([www.randomization.org](http://www.randomization.org)).

#### 105*Supplementation*

106Participants consumed concentrated nitrate rich beetroot juice (containing  $\sim 4.1$  mmol of nitrate; Beet  
107it, James White Drinks Ltd., Ipswich, UK) and nitrate depleted beetroot juice (organic beetroot juice  
108containing  $\sim 0.04$  mmol of nitrate; Beet it, James White Drinks Ltd., Ipswich, UK). Pharmacokinetic data  
109report that plasma nitrate peaks between 2.5 – 3 hours after ingestion of a single dose of beetroot  
110juice (Webb et al., 2008), thus on the day of experimental trials, participants were instructed to  
111consume 70-mL of the supplement 2.5 hours before the beginning of the trial. Both supplements  
112were indistinguishable in taste and smell. Pilot testing with six participants not involved in the main

113study, were unable to identify which supplement had been ingested. The packaging of both  
114supplements were identical in appearance, which were marked by a researcher with a unique code  
115(i.e. “X” or “Y”) for random assignment. One researcher, who was not involved with any experimental  
116testing, knew which codes corresponded to each supplement. To ensure that the placebo blind had  
117been effective, a manipulation check was conducted after each experimental time-trial. Participants  
118were asked to state what supplement they had received by selecting one of three options: 1) beetroot  
119juice; 2) placebo and; 3) don’t know. Participants also indicated what time they had taken the shot, if  
120any habitual practices in training and diet had changed leading up to the trial and if any other factors  
121(e.g. motivation to perform the trial as fast as possible, weather conditions and injuries) affected their  
122performance on the day of the trial.

### 123*Procedure*

124Ethical approval was granted by the lead author’s Institutional Ethics Committee (ref: 14/SAS/189)  
125and parkrun’s Ethics Committee in accordance with the Declaration of Helsinki. Participants were  
126recruited to the study in person and informed about the study’s aim, that participation was voluntary,  
127and that all data collected would be used for research purposes only. After reading the information  
128sheet and completing a health questionnaire, written informed consent was obtained.

129All trials were performed on a Saturday morning at 09:00 at the same location in Kent, United  
130Kingdom between April and May 2015. Ambient conditions were recorded using publicly available  
131data (<https://www.wunderground.com/>) collected by The Weather Company (IBM, Atlanta, Georgia,  
132USA). Minimal differences were reported for all time-trials (temperature =  $11.2 \pm 1.8^{\circ}\text{C}$ ; humidity =  $66$   
133 $\pm 4\%$ ; and windspeed =  $14.6 \pm 2$  km/hr). Participants were instructed to keep exercise and nutritional  
134habits the same, refrain from alcohol 24 hours preceding the trial, high intensity exercise 48-hours  
135prior to the trials and requested not to consume other sport supplements not associated with the  
136study. Participants were instructed to run the 5-km as fast as possible. Trials were performed  
137alongside other runners not involved with the trial. Volunteer parkrun officials recorded completion



138times with data extracted from the official website at a later date (parkrun, 2020). Upon completion,  
139participants reported to the research team who provided instructions for the next trial.

#### 140Data analysis

141Time to complete 5-km for baseline trials were inputted into an online reliability spreadsheet to  
142estimate reliability of pre- and post-baseline trials. Data was log transformed to reduce nonuniform  
143errors and Pearson correlation ( $r$ ), the intraclass correlation (ICC) and CV provided estimates of  
144reliability. The  $r$  coefficient was interpreted as trivial ( $<0.1$ ), small (0.3), moderate (0.5), large (0.7),  
145nearly perfect (0.9) and perfect (1.0; Hopkins, 2015). The ICC was interpreted as low (0.20), moderate  
146(0.50), high (0.75), very high (0.90) and extremely high (0.99; Hopkins, 2015). A paired samples  $t$ -test  
147was conducted to determine systematic differences in performance between baseline trials.

148Data was analysed using SPSS version 24.0 (IBM, Armonk, NY) and tested for homogeneity of variance,  
149normal distribution and outliers. Ratings of supplement assignment (correct, incorrect) were analysed  
150using Chi-square ( $\chi^2$ ). Cramer's  $V$  was used as the effect size and interpreted as 0.10, 0.30 and 0.50,  
151for a small, medium and large effect, respectively (Cohen, 2013). Repeated measures analysis of  
152variance (ANOVA) was conducted to analyse effects of time between conditions. Greenhouse-Geisser  
153epsilon was reported when sphericity was violated. Partial eta-squared ( $\eta^2$ ) is reported as the effect  
154size, with values of 0.02, 0.13 and 0.26 indicating small, medium and large effects respectively (Cohen,  
1551992). Post-hoc Least Significant Difference (LSD) tests were used to examine differences between  
156conditions and Cohen's  $d$  ( $d$ ) was calculated with values 0.2, 0.5 and 0.8 indicating small, medium and  
157large effects, respectively (Cohen, 1992). Data is reported as means  $\pm$  standard error of the mean  
158(SEM) and 95% confidence intervals. Statistical significance was set at  $P < 0.05$ .

#### 159Results

##### 160Preliminary analyses

161 Times were similar between pre- and post-baseline (mean differences =  $16.15 \pm 1.47$  s, 95% CI = -4.80  
162 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  
163 used to measure baseline.

#### 164 *Main analyses*

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

170 Mean times for each condition are shown in figure 1. Repeated measures ANOVA revealed a  
171 significant effect for 5-km time between each condition ( $F_{2, 138} = 13.075$ ,  $P < 0.001$ ,  $\eta^2 = 0.159$ ).  
172 Compared to baseline, participants ran faster in the beetroot (mean differences =  $22.2 \pm 5.0$  s, 95% CI  
173 = 12.2 to 32.1 s  $P < 0.001$ ,  $d = 0.08$ ) and placebo ( $22.9 \pm 4.5$  s, 95% CI = 13.9 to 32.0 s,  $P < 0.001$ ,  $d =$   
174 0.09) conditions. No differences in times were reported between beetroot and placebo ( $0.8 \pm 5.7$  s,  
175 95% CI = -10.6 to 12.1 s,  $P = 0.875$ ,  $d = 0.00$ ).

#### 176 **Discussion**

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

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

1865-km time-trial. These results are similar to Cermak, Res, et al. (2012) and de Castro et al. (2019) who  
187reported that compared to placebo, beetroot juice supplementation does not improve 1-hour cycling  
188time-trial and time to complete 10-km running trial performance, respectively. More recent research  
189(Jodra et al., 2020; Jonvik et al., 2018; Shannon et al., 2017) has reported that beetroot juice is more  
190likely to affect shorter (e.g. 1500-m running) than longer distance (e.g. 10,000-m running) events.  
191Shannon et al. (2017) suggest that dietary nitrate supplementation increases the recruitment of type  
192II muscle fibres and augments blood flow and oxygen delivery. The increase in local blood flow is  
193argued to decrease metabolic perturbations such as PCr degradation and adenosine diphosphate  
194(ADP) accumulation (Vanhatalo et al., 2011), increase muscle force production and ultimately  
195performance (Coggan et al., 2015). Thus, these effects are less likely to impact endurance  
196performance. Given the results of our study, beetroot juice may have little effect on 5-km running  
197time-trial performance.

198The null effects could also be explained by our main outcome variable. To help maximise the validity  
199of our findings, we used an outdoor competitive 5-km time-trial. The physiological effects associated  
200with beetroot juice may not influence performance as much during competitive time-trials than other  
201factors (e.g. social comparisons, rewards for success and anxiety). While a 5-km parkrun may not  
202produce the same psychophysiological response as the Olympics and World Championships, the  
203results of our study are an important first step in identifying whether an acute dose of beetroot juice  
204improves endurance performance in an ecological valid setting. Given that recreational runners  
205arguably account for a substantial proportion of the consumer group for nutritional sport  
206supplements (Maughan et al., 2018), our results highlight that the physiological effects of beetroot  
207juice are unlikely to improve performance for this population. Instead, recreational runners should  
208practice other methods that are more likely to benefit their performance in competitive settings (e.g.  
209an improved training programme, nutritional strategy or psychological profile).

210It is important to consider the reliability of the performance measure when interpreting results. We  
211reported improvements compared to baseline of 1.4% for both the beetroot juice and placebo  
212condition. However, the CV of our measure was 2.66%. It is therefore likely that changes are  
213attributable to systematic and random error. Similarly, the CV of our study is greater than previous  
214research using a similar performance measure (CV = 0.95%; Hurst & Board, 2017). Reasons for the  
215larger variance could be related to the time in-between baseline trials. Hurst and Board (2017)  
216measured 5-km performance twice, separated by 1-week, whereas we separated baseline trials by 3-  
217weeks. Although no differences were shown between baseline trials, it could be speculated that the  
218greater time in-between trials increased the variance in our performance measure. This highlights the  
219importance of measuring a further baseline time-trial after experimental trials to help identify  
220systematic and random error of performance.

221While our performance measure is not as reliable as previous research (Hurst & Board, 2017), the  
222performance measure still holds very good reliability (see Currell & Jeukendrup, 2008). Therefore, the  
223results of our study are supported with high reliability and validity, and a large sample size. Generally,  
224randomised controlled trials in sport and exercise employ small sample sizes and use outcome  
225measurements in tightly controlled laboratories (Burke & Peeling, 2018). This approach can cause  
226difficulties for researchers detecting meaningful changes in performance and translating the findings  
227to applied practice. While challenges exist in recruiting adequate sample sizes and designing studies  
228that are both reliable and valid, the results of this study highlight the opportunity for researchers to  
229analyse the effects of interventions using a reliable and valid measure of running performance with a  
230large sample. By using parkrun as our outcome measure, and recruiting a large sample, this study  
231offers a clearer estimate of the true magnitude of changes in 5-km running performance after  
232administration of an acute dose of beetroot juice.

233*Limitations and future research*

234 While the study has a number of strengths relating to the study design, sample size and outcome  
235 measure, there were limitations. First, we measured the effect of a single acute dose of beetroot juice  
236 (70-mL). There is evidence to suggest that chronic supplementation of beetroot juice may be more  
237 beneficial for improving sport performance than acute supplementation (Jones, 2014; McMahon et  
238 al., 2017). Future research should aim to determine the effect of chronic beetroot juice  
239 supplementation on competitive 5-km running performance. Second, we did not control the content  
240 of nitrate rich foods (e.g. beetroot, lettuce and spinach) in participants' diet. Those with a higher  
241 nitrate rich diet may show reduced effects with beetroot juice supplementation than those with a low  
242 nitrate rich diet (Jones, 2014; Jonvik et al., 2017). Prospective research should consider controlling for  
243 the impact of the consumption of nitrate rich diets in their results. Third, while we recruited a large  
244 sample size that were regular 5-km runners, they were not elite athletes. It is argued that the benefits  
245 of beetroot juice supplementation are more likely to be shown for highly-trained competitive athletes  
246 than recreational athletes due to the consequence of years of training adaptations and genetic factors  
247 (Burke & Peeling, 2018). Future research should aim to sample more highly trained athletes to further  
248 elucidate the effects of beetroot juice on competitive running performance. Fourth, given that our  
249 outcome measure does not mimic the atmosphere, pressure and demands that may be experienced  
250 during competitive events (e.g. national and international championships), and that athletes did not  
251 adjust their training to "peak" for each trial, the "competitive" element of our study is limited. It  
252 would be worthwhile to understand the effects of an acute dose of beetroot juice on running  
253 performance during more competitive events.

#### 254 **Conclusion**

255 In conclusion, our results indicate that there is no difference in competitive 5-km time-trial  
256 performance when participants ingest an acute dose of beetroot juice or an equivalent placebo. This  
257 suggests that beetroot juice may not exert an ergogenic effect on 5-km running performance for  
258 recreational runners. The results of this study are supported with high reliability and validity using a

259large sample size. Future research studies should consider using other parkrun events to investigate  
260the effectiveness of other sport interventions.

**261 Acknowledgments**

262 We would like to thank Dr Katrina Taylor for assistance in the study, parkrun® for granting access to  
263 recruit participants and data, and to the parkrun participants for their involvement in the study.

264

**265 Declarations**

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

267

**268 Authorships**

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

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410Tables

**Table 1 Demographics of participants separated by gender**

	Male	Female	Overall
<i>N</i>	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

411

## 412Figure

413**Figure 1.** Mean time to complete 5-km time-trials for each condition. Data are means  $\pm$  SEM. \* = P

414<0.001 vs. beetroot and placebo.

