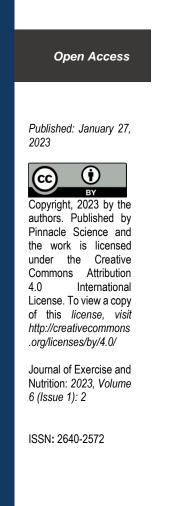


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# Acute Consumption of New Zealand Blackcurrant Extract Has No Effect on Cycling Performance in Normobaric Hypoxia with Trained Cyclists

Original Research

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# Abstract

**Introduction**: New Zealand Blackcurrant Extract (NZBC) is a popular ergogenic aid used to improve endurance performance. The aim of this research was to determine the effects of a single bolus of NZBC on 10-km time trial (TT) cycling performance in normobaric hypoxia.

**Methods:** A double-blind, crossover design study was conducted with trained cyclists. The effects of acute NZBC (900 mg) were compared with a placebo in normobaric hypoxia (NH) (FiO<sub>2</sub> = 15.5%). Testing comprised of three laboratory-based visits for (1) familiarisation (and screening of TT performance before entry into study), (2) placebo and (3) NZBC, whereby a 10-km cycling TT was completed one hour after consumption. After completion of the TT blood lactate was assessed at four time-points in the 10 minutes following. Throughout the TT, power output (PO), rating of perceived exertion (RPE) and heart rate (HR) were recorded.

**Results:** NZBC had no effect on TT cycling performance in NH compared to a placebo (1078.4 s [1009.4, 1147.4] and 1071.0 s [1006.4, 1137.5] respectively, p=0.31; d=-0.31). Additionally, no difference was observed for mean power output (p=0.20; d=0.39), HR (p=0.76; d=0.09) or at 1-km intervals for performance time (p=0.80), PO (p=0.77) or RPE (p=0.41). Post exercise blood lactate recovery did not differ between placebo and NZBC (p=0.42).

**Conclusion**: Acute intake of NZBC has no effect on cycling performance or blood lactate recovery in simulated altitude.

Key Words: Anthocyanins, Exercise Test, Hypoxia

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# Introduction

Research interest in dietary anthocyanins has increased dramatically in the past 10 years <sup>1</sup> with increasing evidence for enhanced sports performance, including improved time trial, repeated sprint and delayed time to exhaustion<sup>2</sup>. Blackcurrants (*Ribes nigrum*) contain naturally high quantities of anthocyanins, which makes them particularly attractive for athletes. Blackcurrant anthocyanins are thought to have anti-inflammatory and anti-oxidant properties, which may reduce oxidative stress during exercise <sup>3</sup> and provide protective effects against mitochondrial defects <sup>4</sup>. Anthocyanin



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concentration in New Zealand blackcurrants are 1.5 times higher than non-New Zealand equivalents with some cultivars presenting concentrations as high as 850 mg per 100 grams of fresh fruit <sup>5</sup>. This makes New Zealand Blackcurrant Extract (NZBC) unique in terms of its properties and suggests an enhanced ability to improve endurance performance.

The reported benefit of blackcurrant anthocyanins on exercise performance is evident across a range of exercise modalities. A recent systematic review reported that NZBC improves sport performance when compared with a placebo (overall effect size = 0.45) during cycling, running and rock climbing performance tests<sup>1</sup>. Effective blackcurrant anthocyanin doses were reported to range between 105-210 mg. However, all studies included in this review implemented a chronic supplementation strategy (7–21 d<sup>-1</sup>). Blood plasma levels of NZBC anthocyanins peak between 1-2 hours following ingestion <sup>6</sup>, suggesting ergogenic effects might be observed immediately following a single, acute consumption of NZBC. A single dose of NZBC (1.87 mg/kg<sup>-1</sup>/BM) is shown to elicit positive effects on forearm blood flow and forearm vascular resistance when sitting but no influence on subsequent hand-grip exercise performance <sup>7</sup>. An increase in blood flow will theoretically increase delivery of oxygen and substrates to muscles, potentially contributing toward enhanced endurance performance <sup>8</sup>. To date, no studies have addressed whether a single bolus of NZBC, taken 1-2 hours prior to exercise influences endurance performance.

Most studies assessing the influence of NZBC on athletic performance have taken place in normoxic conditions. Given that chronic consumption of NZBC increases femoral artery diameter and peripheral blood flow <sup>9,10</sup>, it is plausible that in normobaric hypoxia (NH), performance could be enhanced following acute NZBC intake. Furthermore, when exercising in a hypoxic environment, free radical species are increased beyond that observed in normoxia <sup>11</sup>. Blackcurrant anthocyanins are reported to reduce exercise-induced oxidative stress <sup>3</sup> and this may translate into an attenuation of hypoxia-induced fatigue. Despite this, following 7 days of NZBC consumption at a dose of 600 mg.d<sup>-1</sup>, no difference in substrate oxidation during steady state or 16.1-km cycling performance in NH of 2500m (FiO<sub>2</sub> = 15%) was observed <sup>12</sup>. No other studies have explored the effect of NZBC on performance at altitude. Following four days of blueberry supplementation (anthocyanin = 336 mg.d<sup>-1</sup>), a reduction in blood lactate response during a 30-min running time trial when exposed to normobaric hypoxia (FiO<sub>2</sub> = 15.5%) was reported, however no change in time trial was observed <sup>13</sup>. Furthermore, acute polyphenol supplementation in the form of pomegranate extract resulted in an increased oxygen consumption during a cycling time to exhaustion task at 100%VO<sub>2max</sub> in simulated altitude with highly trained cyclists (FiO<sub>2</sub> = ~17%). Currently, the ergogenic effects of a single dose of NZBC on endurance performance is unknown. The aim of this study was to identify the effects of acute NZBC intake on cycling TT performance in normobaric hypoxia conditions.

#### Scientific Methods

#### Participants

An a priori sample size estimation was performed based on previously observed differences in cycle time trial performance following 7-days consumption of NZBC or a placebo (PLA) <sup>14</sup>. The effect size based on the mean  $\pm$  SD of differences between treatment groups was 0.97. With an alpha=0.05 and power=0.85, the estimated sample size needed to show a statistically significant effect was approximately n=12. Fourteen participants agreed to participate in the study, however two were removed due to listing symptoms of light-headedness during a preliminary 15-min exposure to hypoxic air. Therefore, a total of twelve participants including 8 males ( $24 \pm 4$  years,  $178.2 \pm 3.6$  cm, 79.2 $\pm$  13.8 kg) and 4 females (21  $\pm$  3 years, 167.5  $\pm$  8.4 cm, 70.0  $\pm$  13.9 kg) completed the study. Inclusion criteria were (i) participation in moderate-high intensity physical activity for a minimum of 8 hours per week, (ii) being free of injury or illness for a minimum of two weeks prior to study participation and (iii) completing a 10-km TT cycle in  $\leq 25$ minutes during the familiarisation session. Following familiarisation, participants were screened for 10-km cycling performance (finish time < 25 min) before being entered into the study. All participants were considered trained/developmental (Tier 2) cyclists <sup>15</sup>. Participants were required to complete a 2-day washout out period, abstaining from blackcurrant products and anthocyanin supplements; a duration sufficient to ensure washout of polyphenol presence in urine <sup>16</sup>. During the familiarisation visit, participants were provided with a document listing polyphenol-rich food and drink to avoid prior to the subsequent two visits. In the 24 hours prior to participation, participants were also advised to avoid physical exercise, caffeine and alcohol. Furthermore, participants were requested to consume the same meal and 500 ml of water two hours prior to arrival for all visits. Before beginning the experimental trials, urine osmolarity was measured to confirm euhydration. Participant visits were separated by a minimum of 48 hours with no more than one week between sessions. Ethical approval was received from the University of Essex ethical committee. Participants provided written informed consent in accordance with the Declaration of Helsinki.

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## Protocol

A double-blind, crossover design was implemented to compare the effects of NZBC and a placebo (PLA) during a 10km TT in normobaric hypoxia. At study entry, treatment order was based on non-randomised allocation by presentation (participant 1 = NZBC/placebo; participant 2 = placebo/NZBC). Participants ingested either 900 mg NZBC or a PLA equivalent (gelatine capsules containing 900 mg cornflour). Each NZBC capsule contained 300 mg of active cassis (105mg anthocyanins, 3-10% Cyanidin-3-O-Glucoside, 30-45% Cyanidin-3-O-Rutinoside, 5-20%, Delphinidin-3-O-Glucoside, 35-50% Delphinidin-3-O-Rutinoside, CurraNZ<sup>TM</sup>, Health Currency Ltd., Surrey, UK). All cycling protocols were performed on a Wattbike Pro ergometer (Nottingham, UK). A 5-minute warm up was performed prior to all TT protocols with participants asked to cycle between 70-80 rpm on an air brake resistance of 1. During the time trials initial air brake resistance was set at 3, however participants were free to manipulate the air brake as desired once the TT begun.

Prior to exercise, participants inhaled hypoxic air for 15 minutes produced from an altitude generator set to a FiO<sub>2</sub> of 15.5% (Cloud9, Sporting Edge Ltd., Basingstoke, UK). During this time, participants were also familiarised with the Lake Louise acute mountain sickness (AMS) scale <sup>17</sup> to monitor for detrimental symptoms related to hypoxia exposure. Following the 15-minute hypoxia exposure at rest, participants immediately began a warm-up on the bike prior to undertaking the TT cycle in normobaric hypoxia (NH). During the TT, rating of perceived exertion (RPE) and HR (Polar R300, Kimpele, Finland) were recorded upon completion of each km and the means of each variable were determined prior to analysis.

During the TT, measurements of AMS were taken at 2.5-minute intervals and tissue oxygen saturation (StO<sub>2</sub>, Cloud9 pulse oximeter) measurements were recorded every 0.5-km. Exercise tests were terminated if StO<sub>2</sub> dropped below 75% for longer than 15 seconds. The AMS scale was used for screening and participant welfare purposes only. No exercise tests were terminated because of StO<sub>2</sub> readings. Upon completion of the TT, blood lactate [La] samples were collected at 2.5, 5, 7.5 and 10-minutes post exercise. All blood samples were analysed for [La] within 24 hours (Biosen C-Line Clinic, EKF Diagnostic, Magdeburg, Germany).

#### Statistical Analysis

Central tendency and dispersion of the sample data are represented as mean and 95% confidence intervals (CI). Normality of distribution was assessed using the Shapiro-Wilk test. To determine if an order effect was present, performance time from the first and second visits were analysed with a paired samples t-test. Paired samples t-tests were also used to analyse 10-km performance time, mean PO, HR and StO<sub>2</sub> between conditions (NZBC and PLA). Effect sizes (Cohens d) were used to determine the magnitude of effect between conditions and time points. Cohens d is calculated with PLA as the reference value; therefore, the direction of the effect is observed as either positive or negative (d = PLA - NZBC / pooled standard deviation). A positive effect indicates a higher mean value with PLA vs. NZBC, whereas a negative effect size shows a higher mean value with NZBC. Effect sizes were deemed small (d>0.2), medium (d>0.5) or large (d>0.8)<sup>18</sup>, regardless of the direction of the effect. Individual 1-km 'bins' for performance time, mean PO, HR, RPE and post exercise [La] were analysed using repeated-measures ANOVAs. All data were checked for homogeneity with the Mauchly test of sphericity and if violations were present, adjusted with the Greenhouse–Geiser test. If significant differences were found, the direction of effects were determined using a post hoc Bonferroni correction test. Statistical analyses were completed using SPSS version 28.0 (SPSS Inc., Chicago, Illinois, USA). The level of statistical significance was identified by an alpha value of P < .05.

#### Results

Results indicate no significant order effect for 10-km performance time (p = 0.19). No significant difference was found between NZBC and placebo for mean performance time (p = 0.31), PO (p = 0.20) or HR (p = 0.76) (Table 1). Of the 12 participants, 10 recorded a faster performance time in the placebo condition. Mean tissue oxygen saturation (%) was not different between NZBC (91.2 ± 3.3) and placebo (92.4 ± 3.8) (p = 0.19, d = 0.34). Furthermore, no interaction effects (condition x time) were found for RPE (p = 0.41), power output (p = 0.77), 1-km split performance time (p = 0.80) or [La] at any time point post-exercise (p = 0.42) (Figure 1)

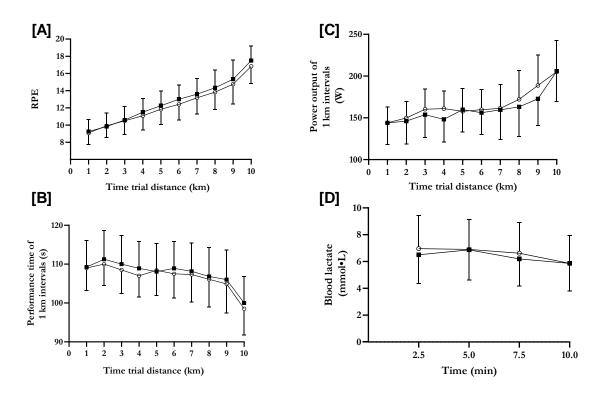


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**Table 1.** Performance and physiological parameters during 10-km time trial in normobaric hypoxia following consumption NZBC and placebo (n=12) (Mean [95% CI])

	NZBC (n = 12)	PLA (n = 12)	95% CI of difference	Effect Size (d)
Performance time (s)	1078.4 [1009.4, 1147.4]	1071.0 [1006.4, 1137.5]	-19.9; 6.9	-0.31
Mean Power (w)	161.0 [133.1, 188.9]	164.7 [138.6, 190.8]	-2.28; 9.63	0.39
Mean HR (bpm)	154 [146, 162]	154 [147, 161]	-3.5; 4.6	0.09

Data are Mean [95% CI]); NZBC = New Zealand Blackcurrant Extract, PLA = Placebo; CI = confidence interval, difference calculated as PLA minus NZBC



**Figure 1.** Mean and 95% CI for (A) Rating of perceived exertion (RPE), (B) performance time, (C) power output at 1-km intervals during the 10-km time trial and (D) post exercise blood lactate in normobaric hypoxia with NZBC ( $\blacksquare$ ) and placebo ( $\circ$ ).

#### Discussion

The aims of this research were to identify whether acute intakes of NZBC had significant effects on TT cycling performance in normobaric hypoxia with trained cyclists. A single 900 mg dose of NZBC produced no effect on performance, HR, RPE or post exercise blood lactate recovery when compared with a placebo. These findings agree with previous research reporting no change in endurance performance in hypoxia, during which 16.1-km cycling time trial was not affected by 7 days consumption of NZBC (600 mg/day active cassis, of which 210 mg/day were anthocyanins), at ~15% FiO<sub>2</sub> <sup>12</sup>. Additionally, no improvement in 30-minute running TT in normobaric hypoxia (15.5% FiO<sub>2</sub>) following four days consumption of blueberry extract (1008 mg/day anthocyanins) was observed <sup>13</sup>. The current study is the first to demonstrate single-dose consumption of NZBC, one hour prior to exercise, has no effect on performance in normobaric hypoxia. Furthermore, peripheral oxygen saturation was not different between NZBC and placebo throughout the TT, indicating that blood flow likely remained unchanged regardless of NZBC consumption when exposed to normobaric hypoxia. Chronic consumption of NZBC has been shown to increase femoral artery diameter, total haemoglobin <sup>10</sup> and peripheral blood flow <sup>9</sup>. It is likely that the acute dose used in this



study did not have similar effects, thus explaining the lack of differences observed. Despite anthocyanin-rich foods appearing rapidly in the blood (1 -2 hours) after consumption, and therefore giving rise to the theory of an ergogenic effect after a single acute dose, the gut microbiota requires a chronic loading period to produce anthocyanin microbial metabolites <sup>6,19</sup>. Chronic consumption of anthocyanin-rich foods is reported to modulate the gut-microbiota composition, resulting in health improvements after a period of weeks rather than days <sup>20,21</sup>. Furthermore, polymorphisms of genes encoding enzymes that contribute to the metabolism of anthocyanins may contribute towards interindividual variation in the microbiota response <sup>22</sup>. Inter-individual response to the consumption of NZBC, including differences in pharmacokinetics of anthocyanin presence in circulation is an emerging area. Recently, phenolic acids found in NZBC were reported to vary in time-to-peak in habitual polyphenol consumers, with protocatechuic acid and gallic acid peaking at 1.5 and 4 hours post consumption respectively <sup>23</sup>. The large interindividual variation that was apparent supports the idea that future research should determine appropriate timings of supplementation prior to exercise at an individualised level.

Various limitations should be considered when interpreting these results. First, diet was not directly controlled, possibly resulting in varied anthocyanin consumption prior to participation. Furthermore, macronutrient consumption may have varied between exercise trails, resulting in varied amounts of muscle glycogen availability. In an attempt to control for this, participants were asked to maintain the same pre-trial dietary intake and avoid products that may contain high quantities of anthocyanins. Given the potential ergogenic effect of a single dose of NZBC, future studies should impose a standardised dietary intake with pre-packaged foods <sup>24</sup>, alongside measuring anthocyanin status pre-trial. Baseline antioxidant status has previously been shown to be an important determinant in the ergogenic potential of an antioxidant supplement <sup>25</sup>. The present study included a combination of male and female participants. However, the phase of the menstrual cycle was not considered. During the late follicular phase, estrogen levels are typically elevated. Estrogen is associated with altering vascular tone, causing vasodilation through nitric oxide pathways <sup>26</sup> and may subsequently mediate endothelial response associated with anthocyanin consumption. Finally, the success of blinding participants was not assessed through post-trial questioning, therefore the influence of expectation bias based on perception of treatment is unknown.

## Conclusions

Acute intake of NZBC had no effect on 10-km TT cycling performance in normobaric hypoxia, suggesting a reduction in FiO<sub>2</sub> presents underlying physiological effects not affected by NZBC intake.

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#### **Conflict of Interest**

No conflict of interest

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