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## Title Page

**Title:** New Zealand blackcurrant extract enhances muscle oxygenation during repeated intermittent forearm muscle contractions in advanced and elite rock climbers

**Running title:** Blackcurrant extract improves muscle oxygen kinetics

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## Abstract

Anthocyanin-rich New Zealand blackcurrant (NZBC) may improve forearm muscle oxygenation and enhance performance in high-level rock climbers. As such, using a double-blind, randomized, cross-over design study, twelve participants performed an oxidative capacity assessment, and two successive exhaustive exercise trials (submaximal forearm muscle contractions at 60 % of their maximal volitional contraction). Each visit was conducted following 7-days intake of 600 mg·day<sup>-1</sup> NZBC extract or placebo. Oxidative capacity was estimated by calculating the oxygen half time recovery using near infrared spectroscopy. Time to exhaustion (s), impulse (kg·s), and minimum tissue saturation index (min-TSI %) were assessed during both the exercise trials. Muscle oxidative capacity was greater with NZBC (mean difference [MD] = 5.3 s, 95% confidence intervals [95% CI] = 0.4 – 10.2 s;  $p = 0.036$ ; Cohen's  $d = 0.94$ ). During the exercise trials, there was an interaction for min-TSI % (time x condition,  $p = 0.046$ ;  $\eta_p^2 = 0.372$ ), which indicated a greater level of oxygen extraction during trial two with NZBC extract (MD = 9 %, 95% CI = 2-15 %) compared to the placebo (MD = 2 %, 95% CI = 1 - 7 %). There was a decrease in time to exhaustion ( $p < 0.001$ ,  $\eta_p^2 = 0.693$ ) and impulse ( $p = 0.001$ ,  $\eta_p^2 = 0.672$ ) in exercise trial two, with no effect of NZBC extract. In high level rock climbers 7-days NZBC extract improves forearm muscle oxygenation with no effect on isolated forearm muscle performance.

**Key Words:** Supplement; nutrition; ergogenic aid; sport climbing; bouldering; rock-climbing

## Introduction

With the rise in the popularity of rock climbing over the past decade, there has been a concomitant increase in both the volume and quality of research that aims to understand the complex and multifaceted physiological components of the sport (D. Giles et al., 2014). Of this research, forearm muscle strength and endurance have been suggested to be some of the most important physical abilities affecting rock climbing performance (Fryer, Giles, Garrido, de la O Puerta, & España-Romero, 2017). For example, the oxidative capacity of the flexor digitorum profundus (FDP) assessed using muscle oxygen time to half recovery ( $O_2HTR$ ) following ischaemia explained 24 % of the variance in climbing performance (Fryer et al., 2016), as well as being able to separate ability groups (Fryer, Stoner, Dickson, et al., 2015), disciplines (Simon Fryer et al., 2017) and arm dominance (D. A. Giles et al., 2016). Given the importance of forearm muscle oxygenation to rock climbing ability, recent studies have attempted to find ways to enhance it using ergogenic aids such as New Zealand black currant (NZBC) extract (Fryer et al., 2020; Potter et al., 2020).

NZBC extract is rich in anthocyanins and has been attributed to improved performance in 16.1. km cycling (Cook, Myers, Blacker, & Willems, 2015), high-intensity treadmill running (Perkins, Vine, Blacker, & Willems, 2015) and rock climbing (Potter et al., 2020). Whilst a comprehensive understanding of the mechanisms involved in these improvements is not yet fully understood, it is thought to be due to an increased blood flow (Potter et al., 2020), likely caused by an altered endothelial function (Speciale, Cimino, Saija, Canali, & Virgili, 2014), potentially upregulating the enzyme endothelial nitric oxide synthase which is involved in the production of endogenous nitric oxide (Suhr, Gehlert, Grau, & Bloch, 2013). As a consequence, blood flow to peripheral muscles would be enhanced by the increased relaxation of smooth muscle cells in the vascular system, and thus vasodilation of the blood vessels (Ziberna, Lunder, Tramer, Drevenšek, & Passamonti, 2013). In support of this potential mechanism, (Matsumoto et al., 2005) found that two hours following blackcurrant intake, forearm blood flow during typewriting was increased. In addition, 7-days intake of anthocyanins has been shown to increase flow mediated dilation of the brachial artery (Khan et

al., 2014; Rodriguez-Mateos, Heiss, Borges, & Crozier, 2014). As such, there appears to be potential for anthocyanin-rich NZBC extract to improve vascular dilator function in rock climbers and thus improve forearm muscle performance.

Only one study has assessed the effects of NZBC extract on brachial artery blood flow and forearm muscle oxygenation in rock climbers (Fryer et al., 2020). Following intermittent forearm contractions at 40 % of maximal volitional contraction (MVC), there were no changes in performance, or macrovascular responses (brachial artery blood flow, diameter, and velocity) however, significant improvements in microvascular function were found. Minimum tissue saturation index (min-TSI %) was lower, and forearm re-oxygenation of TSI % post exercise was quicker with NZBC extract. This improvement in the microvascular response with no change in performance could have been due to i) the low MVC percentage used, or ii) the low ability level of the climbers. Previous research which elicited an improvement in performance variables following a 7-day NZBC supplementation period was conducted using a much higher intensity of exercise (Perkins et al., 2015). In addition, intermediate level climbers, may have been less able to regulate their performance between testing sessions and thus this may have masked any physical differences. As such, the aim of the current study was to determine whether 7-days intake of NZBC extract improved 1) markers of forearm oxidative capacity namely  $O_2HTR$ , and the minimum muscle oxygenation (min-TSI %) achieved during each exercise trial, and 2) performance markers including contraction time (s) and impulse (kg·s) during two exhaustive bouts of isolated forearm muscle contractions in advanced to elite rock climbers.

## **Method**

### *Participants*

The current study used 12 male rock climbers (6 boulderers, 6 sport climbers) classified as advanced (n = 8; 18-23 IRCRA) to elite (n = 4; 24-27 IRCRA) using the grading scale proposed by (Draper et al.,

2016). Participants were excluded from the study if they smoked or were currently taking any vascular acting medication. Participants anthropometric data and climbing characteristics are presented in **Table 1**. Informed consent and health history were confirmed before data collection began, including insuring that they had no recent muscular injuries in the upper limbs or phalangeal joints. Ethical approval from the University of Gloucestershire which conformed to the Declaration of Helsinki (Puri, Suresh, Gogtay, & Thatte, 2009) was obtained prior to participant recruitment and testing.

**Table 1.** Anthropometric and demographic information ( $n = 12$ ).

	Mean	SD
Age (yrs)	25	4
Height (cm)	178	6
Weight (kg)	67.9	5.7
Climbing experience (yrs)	11.5	6.0
Climbing per week (hrs)	5.4	3.3
Training per week (hrs)	5.8	3.1
Climbing ability (IRCRA grade)	23.4	1.9

Yrs = years; cm = centimetre; kg = kilogram; hrs = hours; IRCRA = International Rock Climbing Research Association; SD = standard deviation.

### *Experimental Protocol*

All participants were required to attend the laboratory on three occasions, each at the same time of day. At visit one, participants collected all supplements and performed a familiarisation session on the Digital Lattice Rung. Before visits two and three, all participants were instructed to take 7-days of NZBC or placebo supplement. The order of supplements was allocated using a randomized, counterbalanced, cross-over design (using [www.randomizer.org](http://www.randomizer.org)). Participants were asked not to consume caffeine 6 hours prior to testing, and to avoid light (24 hrs) or strenuous (48 hrs) exercise prior to testing. Participants kept a 48-hour food diary to replicate nutritional eating habits during the loading period before each trial. A small meal, which was kept the same between both laboratory visits, was allowed 2 hours before testing.

During each of the two testing visits participants were weighed upon arrival before completing 20 minutes of passive supine rest, culminating in the forearm oxidative capacity assessment. Following this, participants performed a 15-min self-directed warm-up followed by three MVC trials, a 5-min rest period, and then two successive and exhaustive intermittent fingerboard tests at 60 % of their MVC with a 7:3 s work to rest ratio. For simplicity, from here on these intermittent forearm hangs are described as 'exercise trials'. Each exercise trial was separated by 5-minutes quiet, passive, and seated rest. The TSI % of the FDP was assessed throughout the protocol using NIRS.

#### *Oxidative capacity protocol*

Using a technique that has previously been validated against the standard measurement of phosphocreatine re-synthesis using magnetic resonance spectroscopy (McCully et al., 1994), and used to predict rock climbing performance (Fryer et al., 2016), oxidative capacity was estimated by calculating the O<sub>2</sub>HTR of TSI % using near infrared spectroscopy (NIRS). Briefly, the participants were fitted with a tourniquet (Hokanson Inc, WA, USA) over the brachial artery of the dominant arm. A NIRS optode (Artinis Medical Systems BV, Elst, Netherlands) sampling at 25Hz was then placed over the belly of the FDP in accordance with manufacturer guidelines. The FDP was manually palpated before the location and adipose fat was confirmed using ultrasound (Terrason T3300, Burlington, MA, USA). Adipose tissue was  $1.5 \pm 0.3$  mm deep, well below the  $\geq 6.4$  mm that has been shown to affect NIRS signals (Van Beekvelt, Borghuis, Van Engelen, Wevers, & Colier, 2001). After 20-min quiet supine rest in a temperature-controlled room, participants conducted 10 easy (~10 %) handgrip contractions to activate the metabolism. Immediately post exercise, the tourniquet was inflated to 220 mmHg and sustained until TSI % plateaued at its lowest attainable value for 30 s. Following a stable plateau, the cuff was released and 5-min recovery of muscle TSI % was obtained in order to calculate O<sub>2</sub>HTR. As previously validated (McCully et al., 1994), a reduction in O<sub>2</sub>HTR is concomitant with an increase in skeletal muscle oxidative capacity.



### *Intermittent Forearm Exercise Protocol*

Post warm-up and familiarisation with the testing rung, participants completed two bouts of 7:3 s work to rest ratio hangs at 60 % of their MVC until volitional fatigue, or the participant target force threshold dropped 5 % below target force for more than 2 s. The participant's hand remained on or above the testing rung throughout, and no hand shaking out was allowed as this has been shown to significantly influence muscle oxygen recovery (Baláš et al., 2016). When chalk was needed, it was brought directly up to the hand by a research assistant. Participants received verbal encouragement throughout each trial. Between the two exhaustive exercise trials the participants received 5 min of seated passive rest, during which the hands were placed on their lap and no talking or movement was allowed. Following the final exercise trial, participants were also asked to remain seated for a further 5-min of passive quiet rest. For the exercise trials time to exhaustion (s), mean force (kg) and the impulse (kg·s) were determined for every contraction for all tests; from these variables mean force was calculated across all trial contractions (kg). Time to exhaustion and impulse (kg·s) were defined as total elapsed time up until volitional exhaustion or if force dropped >5 % below target force. The TSI % was assessed throughout the entire exercise protocol with average TSI % representing the mean during each exercise trial, and min-TSI % was determined as an absolute value.

### *Warm-up and familiarisation protocol*

Participants were provided with a 15-min self-directed warm-up in preparation for the finger flexor MVC and intermittent exercise trials. A self-directed warm-up, not prescribed, was used as the highly experienced participants were familiar with warming-up for such exercise. Guidance was provided to split their time between pulse-raising activity, mobilizing, climbing and hangs on the testing edge

(see Figure 1). Participants were also asked to conduct the same warm-up on each of their testing visits to limit any between-day variability. After the warm-up, a re-familiarisation with the Digital Lattice Rung and display was conducted. The familiarisation weight was set at 20 kg and participants were asked to complete four 7:3 s work to rest intervals at this submaximal intensity. This was followed by two 7:60 s work to rest interval hangs at a perceived exertion of ~50 % MVC, and again at ~75 % MVC. Following this, 2 min rest occurred before MVC was determined.

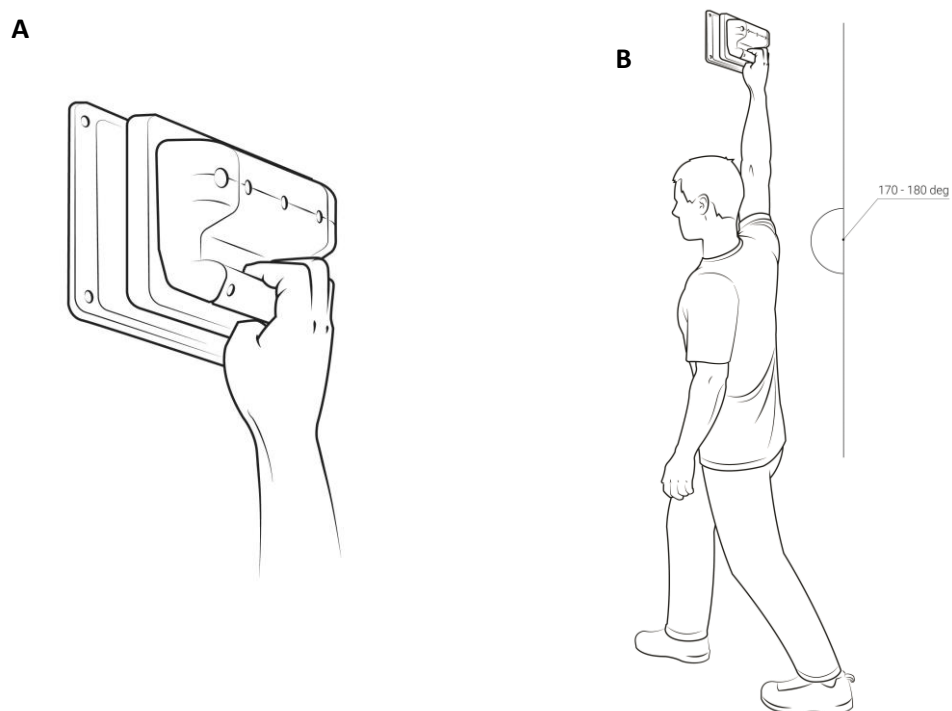
#### *Maximal volitional contraction protocol*

Mean finger flexor MVC was determined on the participant's dominant arm whilst maintaining a 7 s half crimp position 'hanging' on the Digital Lattice Rung. A 7 s average was used as the primary purpose was to judge the intensity for the 7 s contraction repeaters, and this was felt to better represent the contraction intensity at 60 % MVC. Participants were allowed three attempts, with each trial separated by 2-min rest between trials. After MVC was determined, 5-min quiet rest was allowed before the exercise protocol began. Finger flexor MVC, defined as the greatest average force from onset of isometric contraction to beginning of relaxation (kg) from the three attempts, and was determined during both testing sessions with a between visit coefficient of variation of 2.7 %.

#### *Testing rung and hand position*

All participants used a half crimp position on a 20 mm deep and 10 mm radius Digital Lattice Training Rung (Sheffield, England) matching the profile of the testing rung used previously in (D. Giles et al., 2019) and (Torr, Randall, Knowles, Giles, & Atkins, 2020). As shown in **Figure 1**, all tests were performed with the participant's dominant arm extended above the head with 90° flexion at the proximal interphalangeal joint, and a slight bend in the elbow with the shoulder engaged.

Participant's feet remained on the floor and staggered throughout with the leading foot reflecting the dominant arm. When a participant was able to lift their bodyweight off the ground an additional 25 kg ballast was suspended from a climbing harness (loaded under tension). Participants were instructed to develop as much force on the digital rung as possible by 'hanging' (not 'pulling') from the edge. The importance of the distinction became apparent during pilot testing, as pulling result in repeated flexion of the arm and discomfort at the elbow. All finger force data were recorded at a frequency of 80 Hz. Prior to each assessment the digital rung was calibrated and was returned between each test to 0 kg. The data was stored for later offline analysis.



**Figure 1.** Diagram depicting the hand and finger position (A) and participants stance (B) during the exercise trials. Reproduced with permission from Lattice Training Ltd.

#### *Supplementation protocol*

The supplement protocol followed that of (Fryer et al., 2020), (Cook, Myers, Gault, & Willems, 2017), and (Potter et al., 2020) which required participants to consume two capsules each morning for 7 days, and on the final day consume the last two capsules two hours before testing. All capsules were visually identical containing either a placebo of 600 mg microcrystalline cellulose M102 or NZBC

extract of 210 mg anthocyanin (i.e. 35–50 % delphinidin-3-*O*-rutinoside, 5–20% delphinidin-3-*O*-glucoside, 30–45 % cyanidin-3-*O*-rutinoside, 3–10 % cyanidin-3-*O*-glucoside) [CurraNZ<sup>®</sup>, Health Currancy Ltd (Surrey, UK), CurraNZ Ltd (NZ)] per dose of two capsules. A minimum 14-day wash out period was observed between each trial.

#### *Sample size calculation*

Sample size calculations using G\*Power (version 3.1) were based on min-TSI%. With power (1- $\beta$  error probability) set at 0.95,  $\alpha$ -level set at 0.05 and a previously reported (Fryer et al., 2020) effect size of 1.01 (Cohen's *d*), a sample of 9 was required. However, due to the novelty of the repeated measures protocol, and the potential for missing data, we recruited 12 participants.

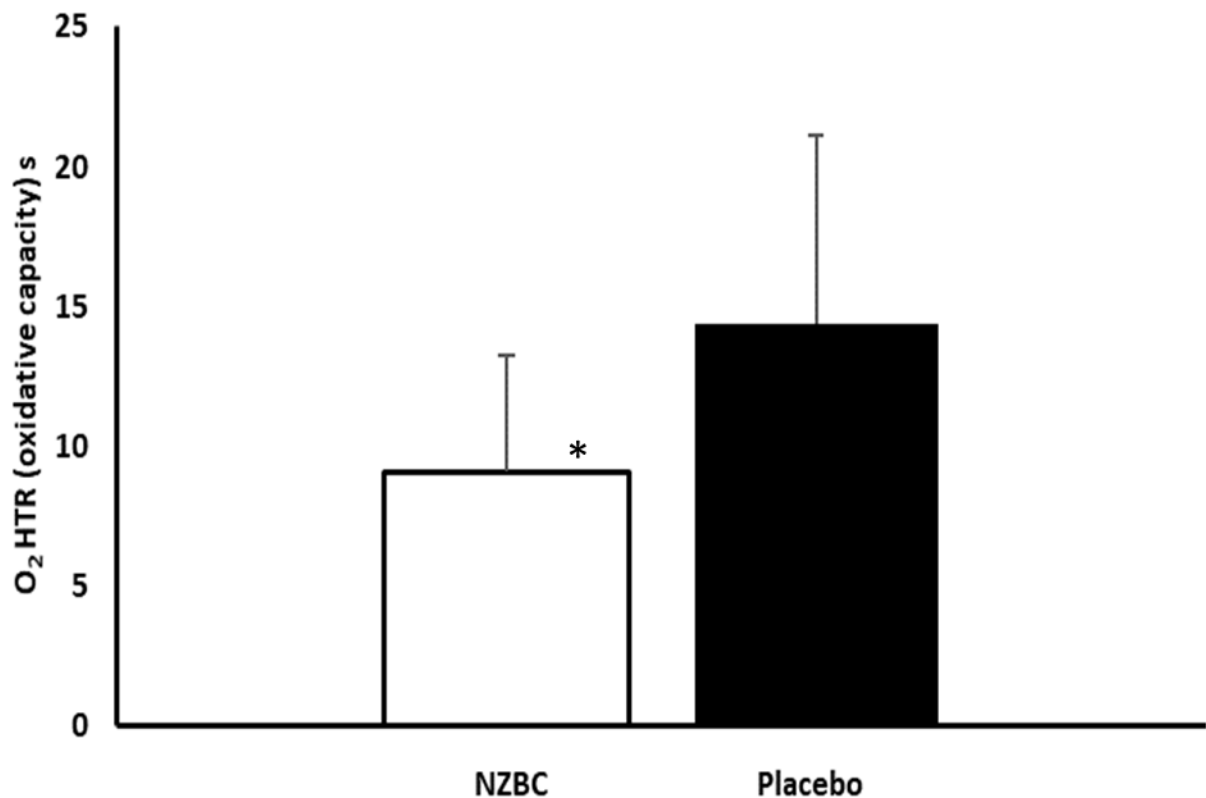
#### *Data analysis*

All statistical analysis was performed using Statistical Packages for Social Sciences (SPSS, Version 25). The  $\alpha$ -level was  $\leq 0.05$ . Normal distribution was confirmed using the Kolmogorov-Smirnov goodness-of-fit test. For all repeated measures data, sphericity was confirmed using Mauchly's test of sphericity. All descriptive data is presented as mean and standard deviation (SD). To determine differences in all dependent variables associated with the fingerboard tests, 2-way repeated measure ANOVAs were used. To assess differences in oxidative capacity between NZBC extract and the placebo trials, a paired samples *t*-test was used. To aid interpretation, mean difference (MD) and 95 % confidence intervals (CI) were determined. Effect sizes for all ANOVA and *t*-test data were calculated using partial Eta<sup>2</sup> ( $\eta_p^2$ ) and Cohen's *d* respectively, where 0.2, 0.5 and 0.8 represent a small, medium and large effect (Cohen, 2013). Due to NIRS equipment malfunction on two participants, all reported muscle oxygenation data is for 10 participants only.

## Results

### *Oxidative Capacity Index (O<sub>2</sub>HTR)*

As shown in **Figure 2**, there was an improved forearm oxidative capacity, as indicated by a 37 % reduction in O<sub>2</sub>HTR, following a 7-day supplement of NZBC extract compared to a placebo ( $9.1 \pm 4.2$  vs.  $14.4 \pm 6.8$  s respectively; MD = 5.3, 95 CI % = 0.4 – 10.2 s;  $p = 0.036$ ,  $d = 0.94$ ) trial.



\* =  $P < 0.05$

**Figure 2.** Mean and SD forearm O<sub>2</sub> Half Time Recovery following a 7-day supplement of NZBC extract and placebo ( $n = 10$ ). NZBC, New Zealand blackcurrant.

### *Hemodynamic data during exercise trials*

Data presented in **Table 2** shows that there was no significant interaction or main effect of time for baseline TSI %. Whilst there was no significant interaction for mean TSI % during the contraction phases of exercise trials one and two. There was a main effect of time with mean TSI % being lower

( $p = 0.005$ ) during the second bout of intermittent contractions compared to the first in the NZBC extract and placebo conditions (MD = 1.9 %, 95% CI = 0.8 – 3.2 %). For min TSI %, there was a significant and meaningful interaction of time x condition ( $p = 0.046$ ). NZBC extract decreased by 8.8 % (95% CI = 2.4 – 15.3 %) between trials one and two whereas the placebo provided a decrease of 1.6 % (95% CI = 1.0 – 7.1 %).

### *Performance data*

A paired samples *t*-test revealed no significant or meaningful differences in MVC or MVC/BW (%) between visits one and two (56.1 vs. 56.2 kg). There were no significant interactions for any of the performance measures presented in **Table 2**. However, there was a main effect of time for both time-to-exhaustion ( $p < 0.001$ ) and impulse ( $p = 0.001$ ). Time to exhaustion decreased between exercise trials one to two in both NZBC extract (MD = 34 s, 95% CI = 17 – 51 s) and placebo (MD = 33 s, 95% CI = 19 – 46 s) conditions. Impulse also decreased from exercise trial one to two in both NZBC extract (MD = 782 kg·s, 95% CI = 381 - 1184 kg·s) and the placebo (MD = 818 kg·s, 95% CI = 455 – 1182 kg·s)

### **Discussion**

The current study aimed to determine the effects of 7-days intake of NZBC extract on the microvascular and performance characteristics in advanced to elite level climbers. Specifically, this study assessed forearm muscle (FDP): 1) oxidative capacity (expressed as  $O_2HTR$ ), and 2) performance responses during two exhaustive intermittent forearm exercise trials at 60% MVC. The main findings of the study are that i) muscle oxidative capacity ( $O_2HTR$ ) assessed following a period of muscle hypoxia, was significantly greater with NZBC extract compared to the placebo trial, ii) during the intermittent contractions to failure, NZBC extract provided a significantly lower min-TSI%

**Table 2.** Mean, SD, interaction of time x supplement, and main effect time for all hemodynamic and performance data during exercise trials one and two following a 7-day NZBC extract and placebo supplementation.

	Exercise Trial 1				Exercise Trial 2				Interaction		Main Effect Time	
	NZBC		Placebo		NZBC		Placebo		Sig.	$\eta_p^2$	Sig.	$\eta_p^2$
	Mean	SD	Mean	SD	Mean	SD	Mean	SD				
<i>Hemodynamic data (n=10)</i>												
Baseline TSI (%)	79.5	5.0	77.0	8.9	79.6	6.1	81.8	3.4	0.114	0.253	0.220	0.162
Mean TSI (%)	42.9	11.8	38.7	9.6	40.9	10.6	36.9	8.6	0.954	0.0001	0.005*	0.604
Min TSI (%)	32.4	10.9	30.0	8.2	23.6	3.3	28.4	5.2	0.046*	0.372	0.015*	0.503
<i>Performance data (n=12)</i>												
Force average over trial (kg)	28.5	3.8	28.8	3.9	28.5	3.9	28.9	4.0	0.828	0.005	0.228	0.129
Time to exhaustion (s)	134.0	23.0	136.6	22.6	100.3	29.2	104.0	24.5	0.809	0.006	<0.001*	0.693
Impulse (kg·s)	2887	789	3047	813	2104	490	2228	498	0.683	0.016	0.001*	0.672

TSI% = tissue saturation index; kg = kilogram; s = seconds; kg·s = kilogram per second; SD = standard deviation;  $\eta_p^2$  = partial Eta<sup>2</sup>; sig = significance; \* =  $p < 0.05$ .

than the placebo trial during both exhaustive exercise trials, iii) there was no effect of NZBC extract on any performance measures during the first exercise trial, and iv) there was a significant decrease in time to exhaustion (s) and impulse (kg·s) between the first and second exercise trials, with no differences between NZBC extract and the placebo conditions.

### *Strengths and limitations*

This was the first known study to assess the effects of 7-days NZBC extract on muscle oxidative capacity, and muscle oxygenation during two maximal exhaustive forearm exercise trials in advanced to elite level climbers. To better contextualise the findings, several strengths and limitations should be highlighted. Firstly, whilst muscle oxidative capacity estimated by calculating the  $O_2HTR$  using cw-NIRS has been previously validated (McCully et al., 1994), it is an estimation and as such may have a small error in comparison to magnetic resonance spectroscopy or muscle biopsy. However, both of these options pose problems as they are expensive and invasive, respectively. Secondly, this study did not assess blood lactate concentration or intramuscular pH, as such the discussion surrounding the effects of muscle acidosis buffering capacity affected by NZBC extract supplementation remains speculative. Lastly, the current study only assessed workload at 60 % MVC during 2 exhaustive trials; previous research which has reported increases in performance with NZBC, has done so using multiple exhaustive trials at higher exercise intensities (Cook et al., 2015; Murphy, Cook, & Willems, 2017; Perkins et al., 2015). Thus, the current study's practical application is limited and more research is needed to better understand the potential effects of NZBC extract during multiple fatiguing protocols at a range of exercise intensities.



### *Comparison with previous studies*

Forearm muscle oxidative capacity was estimated by calculating the O<sub>2</sub>HTR using NIRS (McCully et al., 1994). The current study found that 7-days intake of NZBC extract improved O<sub>2</sub>HTR of the FDP by 5.3 s (Figure 1). Given that a 1 s decrease in O<sub>2</sub>HTR (i.e. an improvement in oxidative capacity) is associated with an increase of 0.7 IRCRA scale (0.7 French sport), and a performance grade of 0.4 (0.4 French sport) separated the top four competitors in the 2015 International Federation Sport Climbing World Cup (Fryer et al., 2016), this finding is an important one. In addition, oxidative capacity of the FDP has previously been seen to separate climbing disciplines (Fryer, Stoner, Dickson, et al., 2015), hand dominance (D. A. Giles et al., 2016), and explain 24 % of variance in overall red-point rock climbing ability (Fryer et al., 2016), and is becoming a useful tool in better understanding the muscle oxygen kinetics and performance of high-level rock climbers. As such it was surprising to find no significant maintenance or improvement of forearm performance with NZBC extract in the current study (**Table 2**). Particularly given the previously suggested importance of oxidative capacity to rock climbing performance, and the fact that NZBC extract has previously been shown to increase time to exhaustion using treadwall climbing (Potter et al., 2020), repeated high intensity running (Perkins et al., 2015), and cycling endurance (Cook et al., 2015). The current study is the second climbing study to report significant changes in muscle oxygenation with no improvement in performance following 7-day NZBC extract supplementation. One potential explanation may be related to the discipline of climbers in the current study. Whilst all were classified as advanced or elite, there were 6 sport climbers and 6 boulderers. Even at 60 % MVC, boulderers would have a greater reliance on local anaerobic strength compared to sport climbers and thus any increase in vessel dilation caused by NZBC may not have been as important for the performance of these individuals. This seems particularly probable given that (Baláš et al., 2016) found that, at least during continuous forearm testing, boulders and sport climbers used different metabolic pathways during their exhaustive exercise bouts at 60 % MVC, whilst achieving the same impulse (Kodejska,

Michailov, & Balas, 2015). Another potential explanation may be related to the nature of the exercise involved. Most previous studies which have reported significant improvements in performance have been conducted with activities which require whole body dynamic movements, unlike the isolated forearm isometric contraction model used in the current study, and that of (Fryer et al., 2020). Whilst the intake of anthocyanins within NZBC extract may result in a dilatory effect within the muscles, this may only be of use for performance when the nature of the exercise requires engagement of multiple muscle groups, and/or a large contribution from the cardiorespiratory system. However, it may also be that whilst O<sub>2</sub>HTR has previously been shown to explain 24 % of the variance in performance (Fryer et al., 2016), it may not actually be as important as initially thought, or that the O<sub>2</sub>HTR is more associated with vessel vasodilation and faster oxygen delivery within the muscle, but not mitochondrial respiration itself. Given the large number of weeks needed to stimulate mitochondrial respiration in training interventions (Granata, Oliveira, Little, Renner, & Bishop, 2016), it would not seem possible that NZBC extract could stimulate improvements in this in 7-days with only 5 hours training. However, this remains speculation and future studies should look to determine the long-term effects of using NZBC during training interventions.

The significantly lower min TSI % without a concomitant improvement in performance seen with NZBC during the second exercise trial is somewhat surprising, and there may be several explanatory factors. Previously, anthocyanins have been shown to increase the perfusion (tHb) within the muscle (Cook et al., 2017), which would result in a greater oxy- deoxyhaemoglobin ratio, and consequently cause a further reduction of the min-TSI %. Further, during exhaustive exercise, more pronounced blood lactate concentrations and greater acidosis may have been observed indicating different substrate utilisation (Perkins 2015; Cook 2015). Intracellular acidosis shifts the dissociation curve of oxyhaemoglobin to the right via the Bohr effect (Grassi, Quaresima, Marconi, Ferrari, & Cerretelli, 1999) resulting in greater capillary O<sub>2</sub> extraction and a decrease in TSI %. One further explanation

may be related to min-TSI % having high biological variability. Baláš, Kodejška, Krupková et al. (2018) found that during intermittent isometric forearm contractions, the reliability of min-TSI % was considered poor (Intraclass Correlation Coefficient = 0.437), as such the measure may not be sensitive enough to account for performance changes in such a homogenous group of climbers. The lower min-TSI % may also relate to the task dependency of muscle fatigue (Enoka, 1995). Both the current study, and that of (Fryer et al., 2020) only used two (at 60 % MVC) and one (at 40 % MVC) exhaustive exercise bouts, lasting up to  $137 \pm 23$  s and  $286 \pm 120$  s respectively. Previous research showing a performance effect of NZBC extract have done so using protocols requiring greater workloads achieved in one or more bouts of exercise, completed at near maximal or supra-maximal work rates (Cook et al., 2015; Murphy et al., 2017; Perkins et al., 2015). It is also plausible that the protocol of the current study may have induced fatigue via different mechanisms than those associated with the higher workloads, such as an accumulation of metabolites and waste products from metabolic pathways, changes in ionic concentrations and a reduced nutrient and energy supply (Girard, Mendez-Villanueva, & Bishop, 2011). Whilst it has been suggested that NZBC extract can postpone peripheral muscle fatigue by allowing elevated levels of intracellular acidosis (Perkins et al., 2015), no known study has assessed this during isolated forearm exercise and thus this remains speculation.

### *Implications*

Research suggests that microvascular rather than macrovascular function is important for rock climbing performance (Fryer et al., 2020; Fryer, Stoner, Scarrott, et al., 2015). Whilst NZBC extract may enhance microvascular function in climbers, more work is needed to examine the potential effect in climbing performance models. However, this may be due to the types of tests used including the intensity, duration, and number of exhaustive contractions. Future studies should look determine the effects of NZBC extract on climbing performance, muscle oxygenation, and

intramuscular pH during multiple exhaustive bouts of exercise at a range of different intensities to further understand the potential mechanisms.

### *Conclusions*

The main findings were that 1) there was a significant improvement in oxidative capacity with 7-day intake of NZBC extract, and 2) whilst min-TSI% was significantly lower during the two exhaustive exercise bouts with NZBC extract, there was no performance effect with repeated intermittent contractions.

### **Conflicts/declaration of interest/disclosure**

There are no conflicts of interests and or relationships between any of the authors and any external partners or companies.

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