INTERNATIONAL JOURNAL OF
SPORT NUTRITION AND
EXERCISE METABOLISM

New Zealand Blackcurrant Increases Post-Exercise Hypotension Following Sustained Moderate Intensity Exercise

Journal:	*International Journal of Sport Nutrition & Exercise Metabolism*
Manuscript ID	IJSNEM.2023-0014.R1
Manuscript Type:	Original Research
Keywords:	New Zealand blackcurrant, anthocyanins, post exercise hypotension, heart rate variability, fat oxidation

ABSTRACT

Previous observations demonstrate New Zealand blackcurrant (NZBC) extract to alter cardiovascular responses at rest without prior exercise. However, the prolonged effects of NZBC on blood pressure and heart rate variability (HRV) following exercise are not known. Participants (*n*15 [5 women], age:31±9 yrs, $\dot{V}O_{2max}$: 44±9 mL·kg^{-1}·min^{-1}) undertook a control condition of 2-hours of laying supine rest. Subsequently, in a double-blind, placebo controlled, randomised cross-over design participants completed 1-hour of treadmill exercise at 50% $\dot{V}O_{2max}$ followed by 2-hour supine rest with blood pressure and HRV measurement following a 7-day intake of NZBC and placebo (PLA). With NZBC, there was an increase in average fat oxidation (NZBC: 0.24±0.11 vs. PLA: 0.17±0.11 g·min^{-1}, *P*=0.005), and larger high frequency relative power during the exercise (P=0.037). In the 2-hour rest period, delta change for systolic pressure was larger with NZBC than placebo (Control vs NZBC: -5.6±6.4, Control vs PLA: -3.5±6.0 mmHg, *P*=0.033), but was not different for diastolic or mean arterial pressure. There were no alterations in HRV variabilities during the 2-hours following the exercise with NZBC. A 7-day intake of NZBC causes a larger post-exercise hypotension response in young, physically active men and women following 1-hour of treadmill exercise at 50% $\dot{V}O_{2max}$.

KEYWORDS: New Zealand blackcurrant; anthocyanins; post exercise hypotension; heart rate variability; fat oxidation

Abbreviations:

DBP Diastolic blood pressure

HRV Heart Rate Variability

MAP Mean arterial pressure

NZBC New Zealand Blackcurrant

PL Placebo

PPAR Peroxisome-proliferator activated receptor

RER Respiratory Exchange Ratio

SBP Systolic blood pressure

$\dot{V}O_{2max}$ maximal rate of oxygen uptake

$_{v}\dot{V}O_{2max}$ Velocity at rate of oxygen uptake

INTRODUCTION

Intake of New Zealand blackcurrant extract (NZBC) has been shown to increase exercise performance (Cook et al. 2019; Braakhuis et al. 2020; Willems and Blacker 2022) and influence cardiovascular responses before (Willems et al. 2015; Cook et al. 2017a) and during exercise (Cook et al. 2017; Cook et al. 2021). Underpinning mechanisms are likely a combination of vasodilation (Cook et al. 2017b; Cook et al. 2021), blood flow (Matsumoto et al. 2005) and increased endothelial nitric oxide synthase (Xu et al. 2004) linked to the antioxidant and anti-inflammatory properties of anthocyanins (Special et al. 2014) and metabolites (Keane et al. 2016).

Willems et al (2015) and Cook et al (2017a) demonstrated NZBC increased cardiac output and decreased total peripheral resistance at rest without prior exercise in trained individuals. Blackcurrant anthocyanins has also been observed to influence

cardiovascular function during exercise. For example, Matsumoto et al. (2005) observed oxygenated haemoglobin in the trapezius to be higher with 2-weeks intake of blackcurrant compared to a placebo during 30-minutes of typing. In addition, maximal voluntary contractions (MVC) of the trapezius performed 3-minutes following the typing, total haemoglobin was also higher. Furthermore, Cook et al. (2017b) observed a 7-day intake of NZBC increased femoral artery diameter during a 120-second isometric contraction at 30% MVC of the knee extensors, with a concomitant decrease in systolic, diastolic and mean atrial blood pressure. Further observations demonstrate that the increased femoral artery diameter during the isometric contraction following NZBC is dependent upon intake duration, with no change from 1-day, but an increase following 4 and 7-days (Cook et al. 2021).

Short-duration, localised cardiovascular effects following exercise with NZBC have also been demonstrated. Following a 7-day intake in rock climbers, Fryer et al. (2020) had participants complete 10 handgrip contractions at ~10% before a tourniquet occlusion of the brachial artery. The study observed a 37% reduction in oxygen half-time recovery in the *flexor digitorum profundus* demonstrating enhanced oxidative capacity.

All these alterations in cardiovascular function, blood flow or oxidative capacity following NZBC intake have been made a few minutes during or following exercise. To the authors knowledge there have been no moderate duration measurements of cardiovascular responses following exercise with NZBC.

Exercise is associated reducing resting blood pressure. The duration and intensity of exercise are important for eliciting prolonged cardiovascular responses following exercise. For example, post-exercise hypotension (PEH) is clinically important due to magnitude of change occurring, with mean systolic and diastolic pressures decreases of 5 and 3 mmHg, respectively, lasting up to 24 hours (Carpio-Rivera et al. 2016). It is caused by multiple physiological responses, including decreased peripheral resistance, sympathetic activity, stroke volume and beta-androgenic receptors and endothelial modulation (Perrier-Melo et al. 2021). It is possible that the influence of NZBC induced vasodilation and blood flow would increase PEH. For example, anthocyanin metabolites have been shown to influence vasoactive properties on vascular smooth cells (Keane et al. 2016) and cause endothelium-dependent relaxation of arteries (Bell et al. 2006).

Heart rate variability (HRV) provides non-invasive analysis on the autonomic influences on the heart. The autonomic nervous system and the relationship between sympathetic and parasympathetic branches and their subsequent contributions to cardiac regulation following exercise with NZBC remain unknown. As sympathetic activity is one mechanism of PEH, it is possible that alterations in blood pressure post exercise by NZBC could be explained by HRV changes. Furthermore, anthocyanins may attenuate cardiovascular disease risk through the mechanisms of regulation of nuclear receptor peroxisome proliferator-activated receptor gamma (Scazzocchio et al. 2011), modulation of nuclear factor-kB (NF-kb) (Stefano et al. 2015) and reducing thrombotic risk (Santhakumar et al. 2013). In turn, there may also be effects of anthocyanins on HRV due to cardiac sympathovagal balance shift toward

parasympathetic activity due to anthocyanin effects upon the microbiome-gut-brain axis (Zong et al. 2023).

To the authors knowledge there have been no studies examining the prolonged cardiovascular responses following exercise with supplementation of NZBC. Therefore, this study aimed to examine the blood pressure and HRV responses in physically active men and women following 1-hour of moderate intensity treadmill exercise following NZBC intake. It was hypothesized that there would be a larger decrease in post-exercise blood pressure following intake of NZBC.

METHODS

Study

The study was approved by the University of Worcester College of Business, Psychology and Sport Research ethics panel (CBPS2122007), with procedures conducted in accordance with the ethical principles outlined by the Declaration of Helsinki (World Medical Association, 2013).

Participants

Fifteen physically active normotensive participants (5 women) volunteered and provided written informed consent, with characteristics presented in Table 1. Participants were health screened and were not smokers or used dietary supplements.

Experimental Design

A double blind, placebo controlled, crossover design was used, with participants visiting the air-conditioned laboratory (20ºC) four times, at the same time of day. Participants abstained from strenuous exercise for 48-hours, alcohol 24-hours before and products containing caffeine on the testing visit days.

124 During visit one, participants underwent screening and had their height (Harpenden
125 Wall Mounted Stadiometer, UK), body mass (Sartorius scales) and blood pressure
126 (Omron M5-I, Omron Healthcare Ltd, Milton Keynes, UK) measured (Table 1).
127 Subsequently, participants completed an incremental treadmill (HP COSMOS,
128 Groningen, Netherlands) protocol to 11 $km \cdot h^{-1}$ with expired gas measurement to
129 determine the linear relationship between running speed and $\dot{V}O_2$. Participants then
130 completed another incremental intensity treadmill protocol to volitional exhaustion to
131 calculate $\dot{V}O_{2max}$. After 20-minutes rest, participants performed a verification square
132 wave protocol, whereby an additional 10% speed added the velocity at $\dot{V}O_{2max}$
133 ($V\dot{V}O_{2max}$). Participants ran at this speed for as long as they could with continuous
134 expired gas measurement (Poole and Jones 2017).
135 In the second visit (control condition), participants rested on a massage plinth for 120-
136 minutes with automated blood pressure measurement on the right arm every 15-
137 minutes.
138 Visits three and four were proceeded by taking two 300 mg NZBC (CurraNZ®,
139 Health Currancy Ltd., Surrey, UK) or placebo (microcrystalline cellulose M102)
140 identical capsules for 6-days prior. For the 6-days, participants consumed one capsule
141 in the morning, and one in the evening, both times with food. On the day of testing
142 (i.e., day-7), participants were instructed to consume both capsules together, 2-hours
143 before arriving at the laboratory in the morning. Each NZBC capsule contained 105
144 mg of anthocyanins. Following randomisation, eight participants received the NZBC
145 condition on visit three. During the debrief of visit four, six participants correctly
146 guessed what experimental condition order they received (40%). There was a 14-day
147 washout between the experimental conditions.

During visits three and four, participants exercised on a treadmill for 60-minutes at 50% $\dot{V}O_{2max}$ with continuous measurement of expired gases (Cortex Metalyzer 3B, Biophysik GmbH, Walter-Köhn-Str. 2d 04356, Leipzig, Germany) and analysis every 5-minutes by averaging the last 60s of breath-by-beath data. Carbohydrate and fat oxidation were calculated using the stoichiometric equations by Jeukendrup and Wallis (2005). Rating of perceived exertion (RPE – Borg 6-20) was measured every 15-minutes. Following the exercise, participants rested on a massage plinth for 120-minutes, with measurement of expired gases for the first 30-minutes and blood pressure every 15-minutes. Participants consumed water *ad libitum*.

Incremental Intensity Walking and Running for Oxygen-speed Relationship

The incremental intensity treadmill protocol in visit one determined the relationship between oxygen uptake and speed. Participants completed 4-minute stages starting at 5 km·h^{-1} and increased by 1 km·h^{-1} until the treadmill reached 11 km·h^{-1} with a 1% incline. Expired gases were averaged for the last 60-seconds of each stage and alongside the $\dot{V}O_{2max}$ and $_{v}\dot{V}O_{2max}$, the running speed eliciting 50% $\dot{V}O_{2max}$ was calculated using linear regression.

Maximal Intensity Treadmill Protocol and Verification of $\dot{V}O_{2max}$

The test commenced at 7 km·h^{-1} for women and 8 km·h^{-1} for men and increased by 1 km·h^{-1} every minute until volitional exhaustion. The square wave verification protocol commenced with a 2-minute period at 7 km·h^{-1} and then abruptly increased to 110% $_{V}\dot{V}O_{2max}$. Participants had no temporal feedback during the protocols but were verbally encouraged. A 15-breath average was used for the highest $\dot{V}O_2$ obtained in both protocols, with the verification stage used to confirm no increase in $\dot{V}O_2$,

despite a higher intensity. There was no increase in $\dot{V}O_2$ obtained in 14 participants in the verification stage (step: 44±9 vs. verification: 42±8 mL·kg^{-1}·min^{-1}, P=0.011).

Blood Pressure

Blood pressure was measured in accordance with methods from the British Hypertension Society. Briefly, participants rested awake, breathing normally without the use of music, books, or electronic devices on a massage plinth with the back rest angled to 45°. The cuff was placed around the upper arm approximately 2 cm above the brachial artery and the artery indicator aligned. Two measurements were taken, and the lowest systolic (SBP) and diastolic (DBP) pressure recorded. Mean arterial pressure (MAP) was calculated by:

MAP = DBP+[(SBP-DBP)÷3]

Heart Rate Variability

HRV during exercise and was recorded continuously using Actiheart 4 (CamNtech Ltd, Cambridgeshire, UK) and analysed on computer software Kubios HRV Standard 3.4.0 (Kubios Oy, Kuopio, Finland). During the 60-minute exercise in visits three and four, HRV was determined from 0-5, 10-15, 25-30, 40-45, and 55-60 minutes. For the 120-minute rest, HRV was determined from 0-5, 10-15, 25-30, 40-45, 55-60, 70-75, 85-90, 100-105, and 115-120 minutes to avoid the blood pressure measurement.

The HRV included time-domain variables, frequency-domain variables, and overview variables. The time-domain parameters were the SD of normal N-N intervals (SDNN) and the root mean square difference of successive normal R-R intervals (RMSSD). Frequency-domain parameters were the absolute and relative power of High Frequency (HF, 0.15-0.40Hz) and Low Frequency (LF, 0.04-0.15Hz) band (APHF, RPHF, APLF, RPLF), Total Power (TP) and the ratio of LF to HF (LF/HF). PNS Index, SNS Index,

and Stress Index were the overview variables analyzed. Beat correction was set to the medium threshold to filter the ectopic beats. Fast Fourier transformation-based Welch's periodogram was performed to calculate the HRV power spectrum.

Statistical Analysis

Statistical analysis was conducted using SPSS 27.0 (IBM SPSS Statistics, Armonk, NY: IBM Corp) and GraphPad 9.4.1 (GraphPad Software, San Diego, CA). Data normality was assessed using Kolmogorov-Smirnov test. Variables were analysed for condition (i.e., NZBC vs. PLA vs. Control), time and interaction effects by a two-way repeated measures ANOVA. Repeated measurements were checked for sphericity using Mauchly's test and if sphericity was violated, Greenhouse-Geisser correction applied. Where differences occurred, subsequent pairwise *post hoc* comparisons were undertaken. Natural logarithm transformation (Ln) was performed on skewed distributed HRV variables before analysis. Delta change (Δ) for the 120-minute average blood pressure changes was also calculated for NZBC and PLA against the control condition. Cohen's *d* effect sizes were calculated (Cohen 1998) with an effect size of <0.2 reported as trivial, 0.2-0.49 as small, 0.5-0.69 as moderate and ≥0.8 as large. Data is presented as mean ± standard deviation.

RESULTS

Metabolic Responses during Exercise and Recovery

Data is analysed from *n*14 due to loss of signal of one participant. Participants exercised on the treadmill at 6.9±1.1 $km \cdot h^{-1}$. During exercise there was no time, condition, or interaction effects for absolute and relative $\dot{V}O_2$, relative intensity, economy, $\dot{V}CO_2$ and heart rate (P>0.05). Minute ventilation demonstrated a time

effect (P<0.001), with no condition or interaction effects (Table 2). RPE demonstrated a time effect (*P*<0.001), with no condition or interaction effect.

Fat oxidation during exercise demonstrated a time (*P*=0.003) and condition effect (*P*=0.005) with no interaction effect (*P*=0.755). Pairwise comparisons indicate differences at 5, (*P*=0.050, *d*=0.46), 10 (*P*=0.008, *d*=0.81) 15 (*P*=0.019, *d*=0.82), 20 (*P*=0.001, *d*=0.84), 25 (*P*=0.032, *d*=0.56), 30 (*P*=0.007, *d*=0.64), 35 (*P*=0.004, *d*=0.63), 55 (*P*=0.048 *d*=0.68) and 60 (*P*=0.044, *d*=0.50) minutes where fat oxidation was higher with NZBC. In addition, average fat oxidation for the 60-minutes was higher with NZBC (NZBC: 0.24±0.11 vs. PLA: 0.17±0.11 g·min^{-1}, *P*=0.005, *d*=0.67), with *n*13 demonstrating an increase. Carbohydrate oxidation also demonstrated a time (*P*=0.002) and condition effect, (*P*=0.028), with no interaction effect (*P*=0.882). Pairwise comparisons indicated carbohydrate oxidation with NZBC was lower at 20 (*P*=0.008, *d*=0.34) and 45 minutes (*P*=0.030, *d*=0.29). Average carbohydrate oxidation was lower with NZBC (NZBC: 1.39±0.42 vs. PLA: 1.50±0.48 g·min^{-1}, *P*=0.027, *d*=0.25). Correspondingly, RER demonstrated a time (*P*<0.001) and condition effect (*P*<0.001) but no interaction effect (*P*=0.134). The pairwise comparisons indicate differences at 5 (*P*=0.029, *d*=0.65), 10 (*P*=0.042, *d*=0.69), 15 (*P*=0.048, *d*=0.71), 20 (*P*=0.027, *d*=0.75), 25 (*P*=0.003, *d*=0.89), 30 (*P*=0.029, *d*=0.45), 35 (*P*=0.004, *d*=0.76) 50 (*P*=0.007, *d*=0.013) and 60 minutes (*P*<0.001, *d*=0.74) (Table 2).

During the 30-minute recovery, there was no condition or interaction effects (*P*>0.05) for absolute and relative $\dot{V}O_2$, $\dot{V}CO_2$, heart rate, minute ventilation, RER, fat oxidation, carbohydrate oxidation and energy expenditure (Supplemental Table 1).

Time effects were observed for absolute $\dot{V}O_2$ (P=0.001), relative $\dot{V}O_2$ (P=0.002), $\dot{V}CO_2$ (P<0.001), heart rate (P<0.001), RER (P<0.001), minute ventilation (P<0.001), fat oxidation (P<0.001) and carbohydrate oxidation (P<0.001).

Recovery Blood Pressure Responses

Resting before the exercise there was no differences between conditions for SBP (Control:113±11, NZBC: 115±13, PLA: 117±12 mmHg, P>0.05), DBP (Control: 69±7, NZBC: 72±5, PLA: 71±6 mmHg, P>0.05) and MAP (Control: 84±7, NZBC: 86±6, PLA: 87±6 mmHg, P>0.05).

For the 120-minute blood pressure measurements, systolic blood pressure demonstrated a time (P=0.002), condition (P=0.002) and interaction effect (P=0.040). At 60-minutes, control was different to NZBC (P<0.001, d=0.62) and placebo (P=0.030, d=0.26), with NZBC also lower than placebo (P<0.001, d=0.34). At 75-minutes, control was different to NZBC (P<0.001, d=0.66) and placebo (P=0.033, d=0.47). At 90-minutes, control was different to NZBC (P<0.001, d=0.94). At 105-minutes, control was different to NZBC (P=0.002, d=0.79) and placebo (P<0.001, d=0.69). At 120-minutes, control was different to NZBC (P=0.010, d=0.63). Delta change for systolic pressure was larger following NZBC than placebo (Control vs NZBC: -5.6±6.4, Control vs PLA: -3.5±6.0 mmHg, P=0.033, d=0.34,).

Diastolic blood pressure was not different for the conditions (P>0.05) but was different across time (P=0.008) with no interaction effect (P>0.05). There was also no difference in delta change for diastolic pressure (Control vs NZBC: -1.6±5.1, Control vs PLA: -0.3±7.0 mmHg, P=0.119). Mean arterial pressure demonstrated no condition effect

(P>0.05) but did have a time effect (P<0.001) and an interaction effect (P=0.043). At 75-minutes, there a was a difference between the conditions (P=0.017), with NZBC lower than the control condition (P=0.010, d=0.57) and the placebo (P=0.044, d=0.36). At 90-minutes there a was a difference between the conditions (P=0.022) with NZBC lower than the control (P=0.007, d=0.69). At 120-minutes there was a strong trend for a difference between the conditions (P=0.054), with NZBC lower than control (P=0.039, d=0.58). Delta change had a trend to be different between the conditions (Control vs NZBC: -2.7±4.6, Control vs PLA: -0.9±5.7 mmHg, P=0.052, d=0.36).

Heart Rate Variability

During exercise high frequency relative power was different between NZBC and placebo (Table 3) (P=0.037), with high frequency relative power larger following NZBC than PLA at 40-45 minutes (P=0.030, d=1.10). There were responses over time for all variables, (P<0.05), however, there was no interactions (P>0.05). Analysed data is from n10 due to signal loss from five participants.

During the recovery period, there was a time response for all the HRV variables (P<0.05), except the low frequency relative power (P=0.217) (Supplemental Table 2). There was no effect condition or interaction effects or for all frequency-domain, time-domain, and overview variables (P>0.05). Analysed data is from n10 due to signal loss from four participants.

DISCUSSION

This study observed that blood pressure decrements after 60-minutes of treadmill exercise at 50% $\dot{V}O_{2max}$ were larger following 7-days of intake of New Zealand

297 Blackcurrant extract in comparison to placebo in healthy, physically active men and
298 women. This was also observed alongside an increase in fat oxidation and decrease in
299 carbohydrate oxidation during the 60-minutes of exercise.
300
301 *Fat Oxidation*
302 The increase in fat oxidation during exercise following NZBC in the present study
303 supports previous observations (Cook et al. 2015; Cook et al. 2017c; Stauss et al.
304 2018; Hiles et al. 2020; Şahin et al. 2021; Şahin et al. 2022; Willems et al. 2022). This
305 study observed an increase in the average fat oxidation of 0.07 $g \cdot min^{-1}$ following
306 NZBC which is similar in absolute increases found by Hiles et al. (2020) of 0.12
307 $g.min^{-1}$, Cook et al. (2015) 0.05 $g.min^{-1}$ and Cook et al. (2017) 0.11 $g.min^{-1}$. The
308 increase in fat oxidation observed in this study was completed in untrained men and
309 women during treadmill exercise at 50% $\dot{V}O_{2max}$ and builds upon previous
310 observations that have used trained individuals, where there would be expected
311 adaptations to increase fat oxidation during exercise. The mechanisms for the altered
312 substrate utilisation are not fully known, however, it is possible the blackcurrant
313 anthocyanins may have effects upon fat metabolism. For example, blackcurrant
314 anthocyanins have been shown to increase mRNA of genes involved with energy
315 expenditure including peroxisome proliferator-activated receptor alpha (PPAR) in
316 C57BL/6J mice (Benn et al. 2014).
317
318 The increase in fat oxidation within this study is a strength as it replicates previous
319 findings and demonstrates that the observations of altered blood pressure post-
320 exercise are also likely a result of the NZBC. Furthermore, this study also adds novel
321 metabolic observations during the 30-minutes following exercise. Interestingly, the

metabolic changes observed during exercise with NZBC is not continued during the immediate recovery. The relative $\dot{V}O_2$ and RER demonstrated time responses within the 30-minute recovery, with both decreasing following the exercise. This is similar to Kuo et al. (2005); however, they demonstrated that the downward trend for RER plateaus at 60-180 minutes following moderate intensity exercise with values lower than before the exercise. Therefore, it is possible that changes with NZBC may be observed at a later duration, rather than during the first 30-minutes of recovery. A limitation to the present study is also that pre-exercise RER was not measured, as a result, the pre to post exercise comparisons in RER and fat oxidation cannot be made.

Blood Pressure Responses and Heart Rate Variability

To the authors knowledge, this is the first study to demonstrate a larger decrease in blood pressure with NZBC in comparison to placebo following exercise. It also supports previous observations of alterations in cardiovascular function with NZBC (Willems et al. 2015; Cook et al. 2017a).

A key consideration when interpreting the findings from this study is that comparisons of post exercise blood pressure were compared to a control condition of no exercise (Figure 1). With the blood pressure measurements following exercise in the NZBC and placebo conditions, there are lots of degrees of freedom, but also comparisons under different conditions, such as rest without prior exercise and rest following exercise. Including a control condition is a strength as it indicates the exercise was sufficient to elicit post exercise hypotension. Furthermore, it also demonstrates that NZBC increases the extent of post-exercise hypotension beyond the post-exercise hypotension observed following the placebo.

347

348 This study measured resting blood pressure in all visits, with the placebo and NZBC
349 measurement made before the exercise. This is another strength as it provided
350 refamiliarisation and also demonstrated that there was no difference in resting blood
351 pressure between the conditions. This was an important measure as post-exercise
352 blood pressure is influenced by the pre-exercise value, where an inflated pre-value
353 could overestimate the extent of post-exercise hypotension (Carpio-Rivera et al.
354 2016). Therefore, this gives further demonstration of post-exercise hypotension.

355

356 In the present study, NZBC decreased systolic blood pressure by 5.6 mmHg in the 2-
357 hours following exercise. This is clinically relevant because a reduction of 3 mmHg in
358 systolic pressure is associated with a 5% reduction in mortality due to cardiovascular
359 disease (Whelton et al. 2002). Furthermore, it is similar to the 5.3 mmHg systolic
360 pressure decrease observed 6-hours following moderate-intensity aerobic exercise in
361 obese individuals by de Lima Bezerra et al (2019) following beetroot juice intake.

362

363 Future research should examine the post-exercise hypotension response with NZBC
364 to identify mechanisms for the response. *In vitro* studies have demonstrated that the
365 anthocyanin cyanidin-3-glucoside can increase the gene expression of nitric oxide
366 synthase (Xu et al. 2004) and enter vascular endothelia smooth cells (Ziberna et al.
367 2012). In addition, using anthocyanin metabolites *in vitro* have also shown migration
368 of vascular smooth cells (Keane et al. 2016). Therefore, it is possible that the
369 anthocyanins from the NZBC increased nitric oxide availability and it turn,
370 vasorelaxation of peripheral arteries.

371

372 The observations from the HRV, however, do not indicate blood pressure changes
373 were caused by alterations in cardiovascular activity (Table 5). To the authors
374 knowledge, this is the first study to measure HRV following NZBC intake. This was
375 completed during exercise and immediately post at rest, and this study found high
376 frequency relative power during exercise with NZBC was larger (Table 3), and low
377 frequency relative power not to change over time during recovery, regardless of
378 condition (Supplement Table 2). Changes in the high frequency band during exercise
379 would suggest alterations in the parasympathetic nervous system. The increase in this
380 study could not solely be interpreted by the increased activation of the
381 parasympathetic nervous system as it can also be influenced by respiratory behaviour
382 (Bae et al. 2021). Furthermore, this study did not observe any changes in RMSSD
383 alongside the high frequency changes, which would have suggested a vagally
384 mediated change in HRV (Shaffer et al. 2017). Future studies should therefore
385 examine the relationship of NZBC, breathing frequency and HRV.
386
387 ***Limitations***
388 Recovery was performed in a seated position on a massage plinth, which is an
389 uncommon method to recover from exercise. Therefore, future research is needed to
390 examine the effects of NZBC on post-exercise hypotension in free-living conditions.
391 Furthermore, blood pressure was recorded in this study for 120-minutes following the
392 exercise and the longer-term effects are not known. For example, reductions in blood
393 pressure have been observed 12.7 hours following exercise in hypertensive
394 individuals (Pescatello et al. 1991) and it is not known if the effects of NZBC on post-
395 exercise hypotension extend to this duration. Furthermore, future experiments
396 examining cardiovascular responses with NZBC should use dietary control, because it

cannot be ruled out that alterations in nitrate (Bailey et al. 2009) or sodium intake influenced blood pressure (Huang et al. 2020).

Conclusions

A 7-day intake of New Zealand Blackcurrant extract increased post-exercise hypotension in comparison to a placebo in the 120-minutes following 60-minutes of treadmill exercise at 50%$\dot{V}O_{2max}$ in physically active men and women. There was also an increase in fat oxidation and decrease in carbohydrate oxidation rate during the exercise, however there was no effect in the immediate 30-minute recovery. There was larger high frequency relative power during exercise with New Zealand Blackcurrant but no effect upon heart rate variability during the recovery.

Author contributions

MDC conceived and planned the experiments. YS and RW carried out the experiments. MDC and YS undertook data analysis. MDC and YS wrote the manuscript.

Acknowledgement

Supply of supplement (CurraNZ™) for this study was obtained from Health Currancy Ltd (United Kingdom) but had no role in any aspect of the study and manuscript. Experiments were performed in the School of Sport and Exercise Science at the University of Worcester (UK). YS and Ruiqi Wang (RW) were supported for this study by the China Scholarship Council. The authors would like to acknowledge RW for assistance in data collection.

Data Availability

Data is available for research purpose upon reasonable request to the corresponding author.

Conflict of interest

The authors report there are no competing interests to declare.

REFERENCES

1. Bae, D., Matthews, J. J. L., Chen, J. J., Mah, L. (2021). Increased exhalation to inhalation ratio during breathing enhances high-frequency heart rate variability in healthy adults. *Psychophysiology*, 58(11), e13905.
2. Bailey, S.J., Winyard, P., Vanhatalo, A., Blackwell, J.R., Dimenna, F.J., Wilkerson, D.P., Tarr, J., Benjamin, N., Jones, A.M. (2009). Dietary nitrate supplementation reduces the O2 cost of low-intensity exercise and enhances tolerance to high-intensity exercise in humans. *Journal of Applied Physiology,* (1985).107(4):1144-55.
3. Bell, D.R., Gochenaur, K. (2006). Direct vasoactive and vasoprotective properties of anthocyanin-rich extracts. *Journal of Applied Physiology*, 100(4):1164-70.
4. Benn, T., Kim, B., Park, Y.K., Wegner, C.J., Harness, E., Nam, T.G., Kim, D.O., Lee, J.S., Lee, J.Y. (2014) Polyphenol-rich blackcurrant extract prevents inflammation in diet-induced obese mice. *The Journal of Nutrional Biochemistry*. 25:1019–1025
5. Braakhuis, A.J., Somerville, V.X., Hurst, R.D. (2020). The effect of New Zealand blackcurrant on sport performance and related biomarkers: a systematic review and meta-analysis. *Journal of the International Society of*

Sports Nutrition, 27;17(1):25. Erratum in: *Journal of the International Society of Sports Nutrition*, (2021) Jan 12;18(1):8.

6. Carpio-Rivera, E., Moncada-Jimenez, J., Salazar-Rojas, W., Solera-Herrera, A. (2016) Acute effects of exercise on blood pressure: A Meta-analytic investigation. *Arquivos Brasileiros de Cardiologia,* 106: 422–433
7. Cohen, J. (1998). Statistical Power Analysis for the Behavioral Sciences, 2nd ed.; Erlbaum Associates: Hillsdale, MI, USA
8. Cook, M.D., Dunne, A., Bosworth, M., Willems, M.E.T. (2021). Effect of Intake Duration of Anthocyanin-Rich New Zealand Blackcurrant Extract on Cardiovascular Responses and Femoral Artery Diameter during Sustained Submaximal Isometric Contraction. *Journal of Dietary Supplements*, 11:1-13.
9. Cook, M.D., Myers, S.D., Blacker, S.D., Willems, M.E. (2015). New Zealand blackcurrant extract improves cycling performance and fat oxidation in cyclists. *European Journal of Applied Physiology*, 115(11):2357-65.
10. Cook, M.D., Myers, S.D., Gault, M.L., Edwards, V.C., Willems, M.E. (2017a) Cardiovascular function during supine rest in endurance-trained males with New Zealand blackcurrant: a dose-response study. *European Journal of Applied Physiology*,117(2):247-254.
11. Cook, M.D., Myers, S.D., Gault, M.L., Edwards, V.C., Willems, M.E.T (2017c). Dose effects of New Zealand blackcurrant on substrate oxidation and physiological responses during prolonged cycling. *European Journal of Applied Physiology*, 117(6):1207-1216.
12. Cook, M.D., Myers, S.D., Gault, M.L., Willems, M.E.T (2017b). Blackcurrant Alters Physiological Responses and Femoral Artery Diameter during Sustained Isometric Contraction. *Nutrients*, 29;9(6):556.

13. Cook, M.D., Willems, M.E.T. (2019). Dietary Anthocyanins: A Review of the Exercise Performance Effects and Related Physiological Responses. *International Journal of Sports Nutrition and Exercise Metabolism*, 1;29(3):322-330.

14. de Lima Bezerra, Á.D., Costa, E.C., Pacheco, D.A., Souza, D.C., Farias-Junior, L.F., Ritti-Dia, R.M., Grigolo, G.B., de Bittencourt Júnior, P.I.H., Krause. M., Fayh, A.P.T. (2019). Effect of Acute Dietary Nitrate Supplementation on the Post-Exercise Ambulatory Blood Pressure in Obese Males: A Randomized, Controlled, Crossover Trial. *Journal of Sports Science and Medicine*, 11;18(1):118-127

15. Fryer, S., Giles, D., Bird, E., Stone, K., Paterson, C., Baláš, J., Willems, M.E.T., Potter, J.A., Perkins, I.C. (2020) New Zealand blackcurrant extract enhances muscle oxygenation during repeated intermittent forearm muscle contractions in advanced and elite rock climbers. *European Journal of Sport Science*, 17:1-9.

16. Hiles, A.M., Flood, T.R., Lee, B.J., Wheeler, L.E.V., Costello, R., Walker, E.F., Ashdown, K.M., Kuennen, M.R., Willems, M.E.T. (2020). Dietary supplementation with New Zealand blackcurrant extract enhances fat oxidation during submaximal exercise in the heat. *Journal of Science and Medicine in Sport*, 23(10):908-912.

17. Huang, L., Trieu, K., Yoshimura, S., Neal, B., Woodward, M., Campbell, N.R.C., Li, Q., Lackland, D.T., Leung, A.A., Anderson, C.A.M., MacGregor, G.A., He, F.J. (2020) Effect of dose and duration of reduction in dietary sodium on blood pressure levels: systematic review and meta-analysis of randomised trials. *The British Medical Journal*. 24;368:m315.

18. Jeukendrup, A.E., Wallis, G.A. (2005). Measurement of substrate oxidation during exercise by means of gas exchange measurements. International Journal of Sports Medicine. 2005;26 Suppl 1:S28-37.

19. Keane, K.M., Bell, P.G., Lodge, J.K., Constantinou, C.L., Jenkinson, S.E., Bass, R., Howatson, G. (2016). Phytochemical uptake following human consumption of Montmorency tart cherry (L. Prunus cerasus) and influence of phenolic acids on vascular smooth muscle cells in vitro. *European Journal of Nutrition*, 55(4):1695-705.

20. Kuo, C.C., Fattor, J.A., Henderson, G.C., Brooks, G.A. (2005). Lipid oxidation in fit young adults during postexercise recovery. *Journal of Applied Physiology,* 99(1):349-56.

21. Matsumoto, H., Takenami, E., Iwasaki-Kurashige, K., Osada, T., Katsumura, T., Hamaoka, T. (2005). Effects of blackcurrant anthocyanin intake on peripheral muscle circulation during typing work in humans. *European Journal of Applied Physiology*, 94(1-2):36-45.

22. Perrier-Melo, R.J., Germano-Soares, A.H., Freitas Brito, A., Vilela Dantas, I., da Cunha Costa, M. (2021). Post-exercise hypotension in response to high-intensity interval exercise: Potential mechanisms. *Revista Portuguesa de Cardiologia (Engl Ed)*, 40(10):797-799.

23. Pescatello, L.S., Fargo, A.E., Leach, C.N. Jr., Scherzer, H.H. (1991). Short-term effect of dynamic exercise on arterial blood pressure. *Circulation*. 83: 1557–1561

24. Poole, D.C., Jones, A.M (2017). Measurement of the maximum oxygen uptake VO2max: VO2peak is no longer acceptable. *Journal of Applied Physiology,* 122: 997-1002.

522 25. Şahin, M.A., Bilgiç, P., Montanari, S., Willems, M.E.T. (2021). Intake
523 Duration of Anthocyanin-Rich New Zealand Blackcurrant Extract Affects
524 Metabolic Responses during Moderate Intensity Walking Exercise in Adult
525 Males. *Journal of Dietary Supplements,* 18(4):406-417.
526 26. Şahin, M.A., Bilgiç, P., Montanari, S., Willems, M.E.T. (2022). Daily and Not
527 Every-Other-Day Intake of Anthocyanin-Rich New Zealand Blackcurrant
528 Extract Alters Substrate Oxidation during Moderate-Intensity Walking in
529 Adult Males. *Journal of Dietary Supplements,* 19(1):49-61.
530 27. Santhakumar, A.B., Bulmer, A.C., Singh, I. (2014). A review of the
531 mechanisms and effectiveness of dietary polyphenols in reducing oxidative
532 stress and thrombotic risk. *Journal of Human Nutrition and Dietetics*. 27(1):1-
533 21.
534 28. Scazzocchio, B., Varì, R., Filesi, C., D'Archivio, M., Santangelo, C.,
535 Giovannini, C., Iacovelli, A., Silecchia, G., Li, Volti G., Galvano, F., Masella,
536 R. (2011). Cyanidin-3-O-β-glucoside and protocatechuic acid exert insulin-
537 like effects by upregulating PPARγ activity in human omental adipocytes.
538 *Diabetes*, 60(9):2234-44.
539 29. Shaffer, F.; Ginsberg, J.P. (2017) An Overview of Heart Rate Variability
540 Metrics and Norms. *Frontiers in Public Health*, 5, 258.
541 30. Speciale, A., Cimino, F., Saija, A., Canali, R., Virgili, F. (2014).
542 Bioavailability and molecular activities of anthocyanins as modulators of
543 endothelial function. *Genes & Nutrition*, 9(4), 404.
544 31. Strauss, J.A., Willems, M.E.T., Shepherd, S.O (2018). New Zealand
545 blackcurrant extract enhances fat oxidation during prolonged cycling in

endurance-trained females. *European Journal of Applied Physiology*, 118(6):1265-1272.

32. Vendrame, S., Klimis-Zacas, D. (2015). Anti-inflammatory effect of anthocyanins via modulation of nuclear factor-κB and mitogen-activated protein kinase signaling cascades. *Nutrition Reviews*. 73(6):348-58.

33. Whelton, P.K., He, J., Appel, L.J., Cutler, J.A., Havas, S., Kotchen, T.A.,Roccella, E.J., Stout, R., Vallbona, C., Winston, M.C., Karimbakas, J. and National High Blood Pressure Education Program Coordinating Committee. (2002) Primary prevention of hypertension: clinical and public health advisory from The National High Blood Pressure Education Program. *The Journal of the American Medical Association*, 288, 1882-1888.

34. Willems, M.E., Myers, S.D., Gault, M.L., Cook, M.D. (2015). Beneficial Physiological Effects with Blackcurrant Intake in Endurance Athletes. *International Journal of Sport Nutrition and Exercise Metabolism,* 25(4):367-74.

35. Willems, M.E.T., Banic, M., Cadden, R., Barnett, L. (2022). Enhanced Walking-Induced Fat Oxidation by New Zealand Blackcurrant Extract Is Body Composition-Dependent in Recreationally Active Adult Females. *Nutrients*. 14, 1475.

36. Willems, M.E.T., Blacker, S.D. (2022) Anthocyanin-Rich Supplementation: Emerging Evidence of Strong Potential for Sport and Exercise Nutrition. *Frontiers in Nutrition,* 31; 9:864323.

37. Xu, J.W., Ikeda, K., Yamori, Y. (2004). Cyanidin-3-glucoside regulates phosphorylation of endothelial nitric oxide synthase. *FEBS Letters,* 10;574(1-3):176-80.

38. Zhong, H., Xu, J., Yang, M., Hussain, M., Liu, X., Feng, F., Guan, R. (2023) Protective Effect of Anthocyanins against Neurodegenerative Diseases through the Microbial-Intestinal-Brain Axis: A Critical Review. *Nutrients*. 18;15(3):496.

39. Ziberna, L., Tramer, F., Moze, S., Vrhovsek, U., Mattivi, F., Passamonti, S. (2012). Transport and bioactivity of cyanidin 3-glucoside into the vascular endothelium. *Free Radical Biology and Medicine,* 1;52(9):1750-9.

Figure 1 A Systolic, B Diastolic, C Mean Arterial Pressure in the control condition of no exercise and 120-minutes following exercise with NZBC or placebo. Data presented as mean±SD. *Control different to NZBC; ‡ Control different to placebo; # NZBC different to placebo.

Table 1. Participant characteristics

Table 2. Volume of oxygen uptake and carbon dioxide produced, relative intensity, heart rate, economy, minute ventilation, carbohydrate, and fat oxidation during 60-minutes of treadmill exercise following placebo and New Zealand Blackcurrant extract.

Table 3. Heart rate variability during 60-minutes of treadmill exercise following placebo and New Zealand Blackcurrant extract.

596 Supplement Table 1. Volume of oxygen uptake and carbon dioxide produced, heart
597 rate, minute ventilation, carbohydrate, and fat oxidation during 30-minutes of rest
598 immediately following 60-minutes of treadmill exercise with placebo and New
599 Zealand Blackcurrant extract.
600
601 Supplement Table 2. Heart rate variability during 120-minutes of rest immediately
602 following 60-minutes of treadmill exercise with placebo and New Zealand
603 Blackcurrant extract.
604

Table 1. Participant characteristics

Age (years)	31±9
Height (cm)	172±10
Body mass (kg)	69±13
$\dot{V}O_{2max}$ ($mL \cdot kg^{-1} \cdot min^{-1}$)	44±9
$\dot{V}O_{2max}$ ($L \cdot min^{-1}$)	3.08±0.84
${}_{V}\dot{V}O_{2max}$ ($km \cdot h^{-1}$)	15±3
RER_{max} (AU)	1.20±0.08
HR_{max}	178±11
Resting Systolic Pressure (mmHg)	117±13
Resting Diastolic Pressure (mmHg)	71±8
Resting Mean Arterial Pressure (mmHg)	86±9
Resting Rate Pressure Product	7.03±1.26

Date reported as mean±SD from 15 participants. $\dot{V}O_{2max}$: maximal oxygen uptake, ${}_{V}\dot{V}O_{2max}$: running speed on treadmill at maximal oxygen uptake, RER_{max}: Maximum Respiratory Exchange Ratio, $HR_{max:}$ maximum heart rate, AU: arbitrary units.

Table 2. Volume of oxygen uptake and carbon dioxide produced, relative intensity, heart rate, running economy, minute ventilation, carbohydrate, and fat oxidation during 60-minutes of treadmill exercise following placebo and New Zealand Blackcurrant extract.

	Time (min)											
	5	10	15	20	25	30	35	40	45	50	55	60
VO_2 ($L \cdot min^{-1}$)												
NZBC	1.56±0.45	1.56±0.46	1.56±0.44	1.55±0.43	1.57±0.45	1.57±0.44	1.57±0.44	1.58±0.46	1.56±0.48	1.58±0.44	1.56±0.44	1.56±0.38
Placebo	1.47±0.41	1.49±0.36	1.53±0.38	1.49±0.39	1.49±0.43	1.51±0.42	1.52±0.42	1.53±0.41	1.55±0.38	1.52±0.38	1.54±0.38	01.56±0.38
VO_2 ($mL \cdot kg^{-1} \cdot min^{-1}$)												
NZBC	22.7±5.0	22.6±5.3	22.7±4.9	22.5±4.8	22.9±5.1	23.1±5.3	22.9±4.9	22.9±5.2	22.6±5.3	22.9±4.9	22.6±4.8	22.6±4.7
Placebo	21.5±5.1	21.7±4.5	22.4±4.6	21.8±4.8	21.6±4.9	22.1±5.0	22.3±5.1	22.4±5.2	22.5±4.6	22.3±4.9	22.5±4.6	22.8±4.8
VCO_2 ($L \cdot min^{-1}$)												
NZBC	1.44±0.42	1.45±0.44	1.47±0.43	1.45±0.43	1.46±0.43	1.45±0.41	1.44±0.40	1.44±0.41	1.42±0.43	1.44±0.40	1.42±0.40	1.41±0.40
Placebo	1.36±0.42	1.41±0.35	1.45±0.43	1.40±0.37	1.39±0.41	1.40±0.40	1.41±0.38	1.41±0.37	1.41±0.36	1.39±0.34	1.41±0.35	1.40±0.34
Relative intensity (%VO_{2max})												
NZBC	49.3±4.2	49.1±2.7	49.4±3.1	48.9±4.1	49.7±3.9	50.2±3.7	49.7±3.6	49.8±3.9	49.1±3.8	49.8±4.0	49.2±3.8	49.1±3.1
Placebo	46.7±3.9	47.3±2.6	48.6±4.1	47.3±4.0	47.0±3.8	48.0±3.6	48.4±3.9	48.7±3.2	48.9±4.1	48.4±4.1	49.0±3.7	49.5±3.2
Heart Rate ($beats \cdot min^{-1}$)												
NZBC	123±15	124±14	124±16	124±16	127±18	128±17	130±20	131±21	131±21	132±21	133±21	132±22
Placebo	123±19	125±16	126±16	126±16	127±16	127±16	128±16	129±16	130±17	131±18	132±18	132±19
Economy ($mL \cdot kg^{-1} \cdot km^{-1}$)												
NZBC	181±40	181±42	182±39	180±39	183±41	185±42	183±40	183±42	181±42	183±37	181±38	181±38
Placebo	171±41	174±42	179±37	174±38	173±39	177±42	178±41	179±39	180±37	180±37	180±38	182±38
VE ($L \cdot min^{-1}$) †												
NZBC	42.0±11.0	44.0±12.6	42.7±10.0	44.0±12.2	45.8±12.7	45.9±13.1	45.9±12.0	45.9±13.5	45.6±13.5	46.4±12.6	46.5±12.7	46.2±12.8
Placebo	42.2±11.4	43.4±10.6	45.0±10.6	44.6±10.4	44.6±12.5	45.4±12.0	45.5±11.5	45.5±11.5	45.1±11.4	45.4±11.1	45.8±11.4	46.5±11.6
Carbohydrate Oxidation ($g \cdot min^{-1}$) †, #												
NZBC	1.31±0.44	1.50±0.52	1.52±0.44	1.42±0.43*	1.44±0.45	1.41±0.55	1.41±0.52	1.37±0.45	1.33±0.44*	1.35±0.39	1.34±0.42	1.25±0.35
Placebo	1.41±0.44	0.56±0.50	1.62±0.54	15.8±0.52	1.57±0.61	1.50±0.50	1.50±0.44	1.48±0.49	1.47±0.51	1.45±0.49	1.44±0.50	1.43±0.52
Fat Oxidation ($g \cdot min^{-1}$) †, #												
NZBC	0.22±0.11*	0.20±0.10*	0.21±0.10*	0.20±0.08*	0.21±0.10*	0.23±0.11*	0.24±0.12*	0.26±0.15	0.27±0.14	0.28±0.20	0.30±0.16*	0.30±0.19*
Placebo	0.17±0.13	0.11±0.10	0.20±0.08	0.13±0.10	0.15±0.11	0.16±0.09	0.17±0.11	0.20±0.12	0.20±0.13	0.20±0.14	0.20±0.13	0.22±0.12
RER (AU) †, #												
NZBC	0.91±0.04*	0.92±0.04*	0.93±0.04*	0.92±0.05*	0.92±0.03*	0.92±0.04*	0.91±0.03*	0.92±0.04	0.92±0.04	0.91±0.04*	0.91±0.04	0.88±0.04*
Placebo	0.93±0.04	0.95±0.03	0.96±0.05	0.95±0.04	0.95±0.04	0.93±0.04	0.94±0.04	0.92±0.04	0.91±0.04	0.93±0.04	0.91±0.04	0.91±0.05

All measures were collected following 7-days of supplementation with NZBC extract or placebo during treadmill exercise. Data reported as mean±SD from 14 participants, †significant effect for time (P<0.05); # significant effect for condition (P<0.05); *denotes (P<0.05) NZBC vs. placebo. AU: arbitrary units.

Table 3. Heart rate variability during 60-minutes of treadmill exercise following placebo and New Zealand Blackcurrant extract.

	Time (min)					ANOVA (P)		
	0-5	10-15	25-30	40-45	55-60	Condition	Time	Interaction
			Frequency Domain					
Low Frequency Relative Power (%)								
NZBC	42.95±14.77	51.84±20.59	55.12±19.24	55.46±18.85	56.38±24.06	0.175	0.003	0.779
Placebo	45.18±15.39	64.62±13.86	63.13±8.59	65.27±13.48	60.53±19.7			
Low Frequency (Ln)								
NZBC	5.6±1.71	4.24±1.39	3.1±1.35	2.87±1.25	2.71±1.43	0.656	0.000	0.912
Placebo	5.24±1.48	3.89±1.77	2.98±1.36	2.81±1.48	2.65±1.23			
High Frequency Relative Power (%)								
NZBC	52.85±16.19	40.94±23.15	34.38±21.64	35.3±20.44*	32.79±24	0.037	0.000	0.702
Placebo	46.26±17.51	25.08±13.44	20.63±11.45	17.81±11.39	26.47±20.43			
High Frequency (Ln)								
NZBC	5.91±1.95	4.05±2.18	2.72±1.78	2.27±1.36	1.87±1.36	0.212	0.000	0.848
Placebo	5.37±1.7	3±2.11	1.92±1.63	1.64±1.43	1.58±1.21			
Low/High Frequency (Ln)								
NZBC	-0.21±0.68	0.32±1.06	0.63±1.1	0.56±0.97	0.67±1.27	0.063	0.000	0.643
Placebo	0.01±0.79	1.03±0.7	1.26±0.71	1.45±0.75	0.98±1.19			
Total Power (Ln)								
NZBC	6.5±1.85	4.98±1.73	3.75±1.54	3.44±1.47	3.28±1.43	0.444	0.000	0.899
Placebo	6.08±1.62	4.35±1.86	3.42±1.39	3.25±1.43	3.08±1.24			
			Time Domain					
SDNN (ms)								
NZBC	46.71±32.02	23.88±25.3	12.25±10.48	9.71±6.24	9±4.95	0.341	0.000	0.783
Placebo	39.53±28.45	15.35±18.8	8.43±5.13	8.08±5.13	7.18±4.27			
RMSSD (ms)								
NZBC	46.83±39.4	31.06±36.33	15.67±17.4	12.16±11.51	10.74±9.3	0.328	0.000	0.811
Placebo	40.88±38.37	17.22±25.96	8.21±4.64	7.88±4.9	8.1±6.22			
			Overview					
Parasympathetic nervous system index								
NZBC	-1.78±1.04	-2.6±1	-3.07±0.74	-3.24±0.54	-3.33±0.49	0.465	0.000	0.659
Placebo	-1.81±0.92	-3.06±0.77	-3.25±0.49	-3.37±0.51	-3.42±0.61			
Sympathetic nervous system index								
NZBC	5.15±1.8	8.04±2.8	10.04±5.89	10.46±5.26	11.07±5.82	0.627	0.000	0.872
Placebo	4.73±2.06	8.61±2.55	11.14±6.74	11.96±6.58	12.78±7.13			
Stress Index								
NZBC	16.95±10.48	31.2±16.59	40.92±25.31	42.02±22.01	44.27±24.63	0.489	0.000	0.735
Placebo	16.76±9.99	32.88±13.54	47.74±28.6	49.98±27.03	52.9±28.36			

All measures were collected following 7-days of supplementation with NZBC extract or placebo during treadmill exercise. Data reported as mean±SD from 10 participants. SDNN; Standard Deviation of the NN intervals, RMSSD; square root of the mean of the sum of the squares of differences between adjacent NN intervals. *different between placebo and NZBC ($P<0.05$).

Supplement Table 1. Volume of oxygen uptake and carbon dioxide produced, heart rate, minute ventilation, carbohydrate, and fat oxidation during 30-minutes of rest immediately following 60-minutes of treadmill exercise with placebo and New Zealand Blackcurrant extract.

	Time (min)					
Condition	5	10	15	20	25	30
VO_2 ($L \cdot min^{-1}$) †						
NZBC	0.32±0.07	0.31±0.08	0.28±0.09	0.30±0.08	0.29±0.06	0.28±0.07
Placebo	0.39±0.10	0.31±0.07	0.30±0.07	0.29±0.08	0.28±0.06	0.28±0.06
VO_2 ($mL \cdot kg^{-1} \cdot min^{-1}$) †						
NZBC	4.69±0.90	4.45±0.74	4.11±1.14	4.32±0.93	4.20±0.79	4.03±0.93
Placebo	4.96±1.16	4.43±1.05	4.28±0.93	4.15±0.72	4.00±0.83	3.94±0.78
VCO_2 ($L \cdot min^{-1}$) †						
NZBC	0.31±0.07	0.28±0.08	0.25±0.08	0.26±0.07	0.25±0.05	0.24±0.06
Placebo	0.34±0.09	0.29±0.06	0.27±0.07	0.26±0.05	0.24±0.05	0.24±0.05
Heart Rate ($beats \cdot min^{-1}$) †						
NZBC	88±34	86±35	82±34	83±36	83±36	80±36
Placebo	84±20	83±21	79±20	81±21	78±22	75±20
VE ($L \cdot min^{-1}$) †						
NZBC	13.63±2.65	12.32±2.35	11.17±2.86	11.45±2.73	10.87±2.09	10.36±2.32
Placebo	15.25±3.75	12.87±2.78	12.12±2.72	11.25±1.80	10.36±2.32	10.33±2.08
Carbohydrate Oxidation ($g \cdot min^{-1}$) †						
NZBC	0.37±0.09	0.28±0.10	0.23±0.10	0.22±0.10	0.18±0.06	0.19±0.09
Placebo	0.40±0.12	0.29±0.08	0.26±0.10	0.21±0.09	0.19±0.09	0.18±0.07
Fat Oxidation ($g \cdot min^{-1}$) †						
NZBC	0.02±0.02	0.05±0.03	0.05±0.02	0.07±0.03	0.07±0.02	0.06±0.06
Placebo	0.02±0.03	0.04±0.02	0.05±0.03	0.07±0.04	0.06±0.03	0.07±0.03
RER (AU) †						
NZBC	0.98±0.07	0.91±0.05	0.89±0.04	0.88±0.05	0.85±0.03	0.87±0.02
Placebo	0.97±0.07	0.93±0.06	0.91±0.07	0.87±0.09	0.87±0.07	0.86±0.06

All measures were collected following 7-days of supplementation with NZBC extract or placebo during 30 minutes of rest following 60 minutes treadmill exercise. Data reported as mean±SD from 14 participants, †significant effect for time (P<0.05).

Supplement Table 2. Heart rate variability during 120-minutes of rest immediately following 60-minutes of treadmill exercise with placebo and New Zealand Blackcurrant extract.

	Time (min)									ANOVA (P)		
	0-5	10-15	25-30	40-45	55-60	70-75	85-90	100-105	115-120	Condition	Time	Interaction
Frequency Domain												
Low Frequency Relative Power (%)												
NZBC	51.25±12.79	45.15±13.43	44.84±17.43	52.19±22.18	55.55±15.65	49.58±18.34	51.43±21.62	53.74±16.93	55.35±16.89	0.764	0.217	0.224
Placebo	61.14±14.98	58.56±20.08	45.49±16.45	47.53±20.58	54.59±19.39	51.21±18.64	49.79±16.06	53.14±15.34	55.81±19.18			
Low Frequency (Ln)												
NZBC	6.28±0.88	6.55±0.78	6.98±0.9	7.08±0.87	7.16±0.92	7.02±1.01	6.96±0.97	7.29±0.67	7.37±0.51	0.970	0.000	0.667
Placebo	6.21±1.33	6.94±1.03	6.79±1.05	6.74±0.97	7.29±0.69	7.19±0.69	7.11±0.8	7.36±0.43	7.47±0.73			
High Frequency Relative Power (%)												
NZBC	40.9±13.61	49.59±14.15	49.72±19.49	40.56±23.66	34.12±16.55	41.02±22.02	42.12±24.38	32.68±17.83	36.56±17.43	0.803	0.007	0.091
Placebo	29.27±14.39	35.28±19.21	47.6±19.65	42.69±22.9	38.36±20.24	36.61±22.84	42.99±16.28	38.04±17.14	37.73±20.46			
High Frequency (Ln)												
NZBC	6.01±1.14	6.62±0.7	7.11±0.8	6.77±0.89	6.61±0.98	6.78±1.02	6.77±1.14	6.7±0.92	6.93±0.85	0.841	0.000	0.428
Placebo	5.38±1.66	6.34±0.95	6.8±1.08	6.57±1.03	6.87±0.55	6.78±0.74	6.98±0.74	6.97±0.61	7.08±0.74			
Low/High Frequency Ratio (Ln)												
NZBC	0.49±0.97	0.13±0.91	0.13±1.2	0.48±1.23	0.67±0.83	0.37±1.09	0.38±1.42	0.74±1.01	0.56±0.89	0.810	0.036	0.140
Placebo	0.97±0.87	0.81±1.2	0.19±1.07	0.34±1.18	0.55±1.06	0.55±1.13	0.33±0.96	0.59±1.03	0.49±1.07			
Total Power (Ln)												
NZBC	6.98±0.93	7.38±0.67	7.88±0.69	7.85±0.68	7.79±0.88	7.82±0.87	7.8±0.78	7.96±0.62	8.05±0.55	0.908	0.000	0.678
Placebo	6.73±1.39	7.54±0.82	7.64±0.93	7.58±0.77	7.97±0.31	7.95±0.46	7.88±0.66	8.05±0.31	8.17±0.49			
Time Domain												
SDNN (ms)												
NZBC	34.34±11.71	45.49±14.7	56.99±19.55	53.12±17.71	53.62±19.63	57.36±22.73	58.81±20.89	61.19±19.47	62.06±16.49	0.820	0.000	0.873
Placebo	32.13±16.26	46.53±18.16	53.43±23.71	49.19±18.04	56.75±11.7	56.71±13.73	55.93±16.62	58.68±14.6	62.57±16.61			
RMSSD (ms)												
NZBC	31.44±13.94	49.15±22.86	64.16±31.8	56.1±29.11	54.18±24.26	62.23±31.53	62.52±30.31	59.16±25.68	61.06±21.91	0.867	0.000	0.608
Placebo	27.37±14.61	46.4±21.48	57.29±33.05	51.4±23.4	58.79±15.2	58.55±20.37	61.77±21.16	60.56±21.66	65.04±25.49			
Overview												
Parasympathetic nervous system index												
NZBC	-1.36±0.84	-0.04±1.34	0.65±1.59	0.58±1.52	0.29±1.25	0.93±1.56	0.91±1.45	0.7±1.24	0.87±1.32	0.942	0.000	0.198
Placebo	-1.7±0.73	-0.23±1.02	0.39±1.47	0.35±1.04	0.77±0.86	0.76±0.98	1.05±0.95	0.96±1.13	1.11±1.14			
Sympathetic nervous system index												
NZBC	2.05±1.32	0.45±1.39	-0.14±1.41	-0.21±1.27	-0.1±1.22	-0.53±1.25	-0.59±1.09	-0.48±0.98	-0.58±1.1	0.933	0.000	0.616
Placebo	2.71±1.66	0.64±1.52	0.08±1.28	-0.09±0.86	-0.61±0.53	-0.6±0.54	-0.76±0.62	-0.74±0.7	-0.82±0.64			
Stress Index												
NZBC	12.94±3.54	10.08±3.83	8.44±3.78	8.88±3.3	7.75±4.65	7.94±3.55	7.48±2.6	7.61±2.65	7.35±2.4	0.762	0.000	0.773
Placebo	14.46±5.13	10.83±5.69	9.53±4.3	9.42±3.21	7.47±1.09	7.54±1.3	7.64±1.93	7.36±1.64	6.99±1.5			

All measures were collected following 7-days of supplementation with NZBC extract or placebo following 60-minutes of treadmill exercise. Data reported as mean±SD from 10 participants. SDNN; Standard Deviation of the NN intervals, RMSSD; square root of the mean of the sum of the squares of differences between adjacent NN intervals.

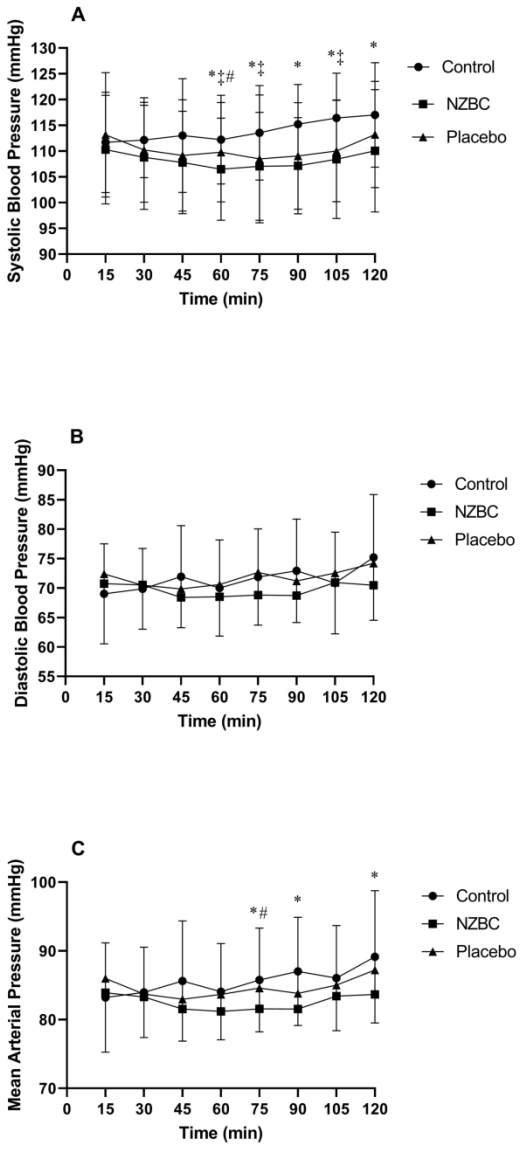

Figure 1 A Systolic, B Diastolic, C Mean Arterial Pressure in the control condition of no exercise and 120-minutes following exercise with NZBC or placebo. Data presented as mean±SD. *Control different to NZBC; ‡ Control different to placebo; # NZBC different to placebo.

127x266mm (300 x 300 DPI)