

1 Title: Effects of New Zealand blackcurrant extract on exercising substrate utilisation and
2 post exercise blood pressure in males and females.

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18 Running Title: Blackcurrant effects in males and females

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25 **Abbreviations:**26 ΔCHOOX delta change of carbohydrate oxidation27 ΔFATOX delta change of fat oxidation

28 DBP diastolic blood pressure

29 MAP mean arterial pressure

30 NZBC New Zealand blackcurrant

31 **FATOX** **Fat oxidation**

32 PLA placebo

33 RER respiratory exchange ratio

34 SBP systolic blood pressure

35 $\dot{V}\text{O}_{2\text{max}}$ maximal rate of oxygen uptake36 $v\dot{V}\text{O}_{2\text{max}}$ velocity at rate of oxygen uptake

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45 **ABSTRACT**

46 New Zealand blackcurrant extract (NZBC) has been shown to increase fat oxidation during
47 exercise and the post-exercise **blood pressure** in males and females. The change in fat
48 oxidation by NZBC has also been shown to be correlated to body composition in males and
49 females. There has never been a comparison of sex responses within the same study. Twenty-
50 two participants (11 males and 11 females, age: 29 ± 8 years, maximal oxygen **uptake**
51 ($\dot{V}O_{2max}$): 44 ± 9 mL·kg⁻¹·min⁻¹, body fat: 18 ± 6 %) had resting blood pressure measured for 2
52 hr (**no exercise**). In a double-blind, placebo-controlled (PLA), randomised crossover design,
53 participants completed one hour of treadmill exercise at 50% $\dot{V}O_{2max}$ with expired gas
54 measurement, followed by 2-hr **resting** blood pressure measurement **with 7-days of NZBC or**
55 **PLA**. Average fat oxidation was different between the conditions (NZBC: 0.27 ± 0.11 , PLA:
56 0.21 ± 0.12 g·min⁻¹, $P < 0.001$), but the response between males and females was not different.
57 When combined, there was no relationship ($P > 0.05$) between body fat percentage **and** change
58 in fat oxidation ($\Delta FATOX$) ($r = -0.079$), with males also demonstrating no relationship ($r =$
59 0.069), but females did demonstrate a relationship ($r = 0.691$, $P < 0.05$). In the 2-hr rest, systolic
60 pressure delta change was larger with NZBC than PLA (no exercise vs. NZBC: -5.5 ± 5.4 , vs.
61 no exercise vs. PLA: -2.9 ± 5.1 mmHg, $P < 0.001$) but was not different between males and
62 females. **A 7-day intake of NZBC extract increases fat oxidation during moderate-intensity**
63 **exercise and decreases post-exercise blood pressure in males and females. The magnitude of**
64 **change in fat oxidation in females is correlated to body fat percentage.**

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66 **Key words:** anthocyanins, fat oxidation, blood pressure

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69 INTRODUCTION

70 Blackcurrant is a fruit with a high content (~590 mg/100g) of the flavonoid anthocyanin,
71 mostly derived from the anthocyanidins delphinidin (66.1%) and cyanidin (31.6%) in
72 glucoside and rutinoside forms (Rothwell et al., 2013). Anthocyanins give dark pigmentation
73 to fruits with the amount affected by ultraviolet light (Guo et al., 2008), with recent studies
74 examining exercise performance and physiological effects following New Zealand
75 blackcurrant (NZBC) extract (Cook et al 2015; Şahin et al., 2022).

76 Anthocyanin confers health benefits partly by anti-inflammatory and anti-oxidant effects
77 (Cerletti et al., 2017; Cappellini et al., 2021). This likely results from the high delphinidin
78 content, which of all the anthocyanins has the highest number of hydroxyl groups and B-ring
79 located hydroxyl groups which confer intracellular radical scavenging (Yi et al 2010).

80 Furthermore, under conditions where cell oxidative stress and inflammation occur such as
81 high intensity exercise, this may also explain that blackcurrant extract supplementation has
82 been shown to increase exercise performance (see reviews: Braakhuis et al., 2020; Cook and
83 Willems 2019; Willems and Blacker 2022). In addition, studies have also shown positive
84 responses from NZBC extract that would support exercise performance such as altered
85 cardiovascular function (Willems et al., 2015) and substrate utilisation (Cook et al., 2015).

86 Despite the radical scavenging ability of the parent anthocyanins, it is likely that metabolites
87 are the main causes due to higher bioavailability and synergistic activity (Keane et al 2016).

88 The ability to oxidise fat during exercise is important for exercise performance. During sports
89 where the use of limited stored energy substrates is important, increasing fat oxidation
90 (FATOX) could be important to preserve endogenous carbohydrate stores. For example, the
91 ability to use fat during exercise is positively associated with performance in an ironman
92 triathlon in men (Frandsen et al., 2017) and women (Vest et al., 2018). Furthermore, body

93 composition also influences fat oxidation during exercise, with a higher body fat percentage
94 increasing utilisation of fat (Goodpaster et al 2002).

95 Increased **FATOX** has been observed after 7-days intake of **NZBC** extract intake. For
96 example, Cook et al. (2015) observed in trained male cyclists a 27% increase in **FATOX**
97 (Placebo:0.37±0.15, NZBC:0.44±0.12 g·min⁻¹) during 10-minutes of ergometer cycling at
98 65% $\dot{V}O_{2max}$ following 105 mg·day⁻¹ anthocyanins. Furthermore, during 120-minutes
99 ergometer cycling at 65% $\dot{V}O_{2max}$, intake of 210 mg·day⁻¹ of anthocyanins increased
100 **FATOX** by 22% in males (Control:0.63±0.2, NZBC:0.74±0.13 g·min⁻¹), while Strauss et
101 al. (2018) observed a 27% increase in females for same duration and intensity of cycling
102 (Placebo: 0.34±0.1, NZBC: 0.40±0.07 g·min⁻¹).

103 **The** magnitude of change in **FATOX** following NZBC extract intake may be associated with
104 body fat percentage. For example, Şahin et al. (2022) demonstrated a correlation between
105 body mass index and body fat % and the absolute change in **FATOX** ($R^2=0.3968$, $R^2=0.5662$,
106 respectively) during moderate intensity treadmill walking following 14-days NZBC intake in
107 recreationally active males. Similarly, Willems et al. (2022) observed that intake of 600 mg
108 of NZBC extract capsules (210 mg anthocyanin) for 7-days to increase **FATOX** during
109 moderate intensity (4.7±0.4 MET) treadmill walking in recreationally active females, with the
110 absolute change over placebo correlated to body mass index ($R^2=0.53$) and body fat
111 percentage of the legs ($R^2=0.57$), arms ($R^2=0.46$) and trunk ($R^2=0.44$). **However, both**
112 **experiments only had single sex samples, and comparisons between males and females**
113 **following NZBC intake within the same study have not been investigated. Therefore, it is**
114 **unclear if there are effects of sex and body composition in substrate utilisation responses**
115 **following NZBC extract.**

116 NZBC extract has also **influenced** cardiovascular responses during exercise by increasing
117 femoral artery diameter (Cook et al., 2017) and increasing **the** post-exercise hypotension
118 **response** (Shan and Cook 2023). **Similarly**, the cardiovascular responses following NZBC
119 extract intake between males and females have never been compared. **It is not known if**
120 **different responses could occur, especially as females and males have been shown to have**
121 **similar post exercise blood pressure responses, however the hemodynamic determinants are**
122 **different, with males having a decrease in cardiac output, while in females there is a decrease**
123 **in systemic vascular resistance (Queirox et al 2013). As a result, a male versus female**
124 **comparison is important to inform guidance on intake, and characteristics that could have a**
125 **greater response to NZBC extract intake.**

126 **Furthermore**, the responses to NZBC extract for males and females may be different. For
127 example, following absorption, polyphenols undergo monooxygenation and are then
128 catalysed by phase II enzymes. Isoforms of these enzymes have differing activity in males
129 and females (Campesi et al., 2018). Furthermore, there have been observations of sex
130 differences in the efficacy of berry polyphenols to reduce oxidative stress. For example,
131 oxidized low-density lipoproteins decreased to a greater extent following a high fat meal
132 challenge in females compared with males (Female: -4.6 ± 2.7 , Male: -3.2 ± 3.0 U/L)
133 following 6-weeks of daily 10 g freeze-dried strawberry beverage consumption (~338 mg
134 phenolic compounds - **quercetin, ellagic acid, anthocyanins, catechins, and kaempferol**)
135 (Burton-Freeman et al., 2010) and 4-weeks of the polyphenol genistein, (600 mg/kg food) **an**
136 **isoflavone**, was shown to reduce systolic blood pressure in female murine only (Al-Nakkash
137 et al., 2012).

138 Elliot-Sale et al. (2021) highlighted the importance of including females into research to
139 inform sex-specific guidelines. In addition, Smith et al. (2022) identified in an audit of sports
140 nutrition research that fewer than 4% of studies conducted comparison of males versus

141 females within the same study. To the author's knowledge, there have been no male versus
142 female comparisons **in one study** on physiological, metabolic, and cardiovascular responses
143 to the intake of NZBC extract.

144 The aim of this experiment was to compare metabolic responses during moderate intensity
145 treadmill exercise following NZBC **extract** intake between males and females, and further
146 examine if post-exercise blood pressure responses were similar. The study hypothesised that
147 post-exercise blood pressure responses between males and females would be similar.
148 Furthermore, NZBC extract would increase **FATOX**, but the absolute change would be
149 similarly correlated to body fat percentage in males and females.

150 **METHODS**

151 **Ethical approval**

152 Ethical approval was obtained from the University of Worcester's College of Business,
153 Psychology and Sport Research ethics panel (CBPS2122007), with procedures conducted in
154 accordance with the ethical principles outlined by the Declaration of Helsinki (World
155 Medical Association, 2013).

156 **Participants**

157 Twenty-two **self-reported** physically active participants (11 females) volunteered, **were health**
158 **screened** and provided written informed consent **with characteristics presented in Table 1. To**
159 **participate, participants had to not be using any dietary supplements, non-smokers, not obese**
160 **(BMI \leq 30), and free from any metabolic health disorders. Female participants were allowed to**
161 **continue with contraception during the study – with 7 reporting use. Participants were also**
162 **categorised as high-normal or below (Table 1) and confirmed in visit one from the right arm**
163 **with no participant achieving a resting blood pressure of \geq 140 mmHg systolic and \geq 90**
164 **mmHg diastolic (Williams et al 2018). The same arm was used for all blood pressure**

165 measurement in the remaining visits. **Participants were excluded if they had cardiovascular**
166 **disease, were taking medication to control blood pressure, or had musculoskeletal injuries**
167 **that would be exacerbated by the testing.**

168 *Table 1 here.*

169 **Experimental Design**

170 The experimental design has previously been described (Shan and Cook 2023). A double-
171 blind, placebo-controlled, randomised cross-over design was used with four laboratory visits
172 at the same time **in the morning**. The air-conditioned laboratory was maintained at 20°C.
173 Prior to all visits, **refrained** from exercise for 48-hours, alcohol for 24-hours and caffeine on
174 the day of the testing.

175 During visit one, height (Harpenden Wall Mounted Stadiometer, UK), body mass (Sartorius
176 scales), blood pressure (Omron M5-I, Omron Healthcare Ltd, Milton Keynes, UK) and body
177 composition (Bodystat 1500, Bodystat Ltd, Douglas, Isle of Man) were measured.

178 Participants then completed an incremental motorised treadmill protocol (HP COSMOS,
179 Groningen, Netherlands) to 11 km·hr⁻¹ with expired gas measurement to determine the linear
180 relationship between running speed and $\dot{V}O_2$. **After a 5-minute rest**, this was followed by a
181 second incremental intensity protocol until voluntary termination to calculate $\dot{V}O_{2max}$.

182 Following a 20-minute rest, a square wave verification protocol was performed at 110%
183 velocity at $\dot{V}O_{2max}$ ($v\dot{V}O_{2max}$) until voluntary termination (Poole and Jones 2017).

184 In the second visit (**i.e., no exercise**), participants rested on a massage plinth for 120-minutes
185 with blood pressure measured every 15-minutes. **Visits one and two and were separated by**
186 **72-hours. While visits two to three were separated by a minimum of 7-days to allow for**
187 **consumption of the NZBC extract or placebo.**

188 For 6-days prior to visits three and four, participants consumed either NZBC (CurraNZ®,
189 Health Currancy Ltd., Surrey, UK) or placebo (microcrystalline cellulose M102) from two
190 300 mg identical size, shape, and colour capsules. A dose-response study indicated 600 mg
191 for 7-days reduces resting mean arterial pressure in endurance trained men, with no additional
192 benefit of a higher dose (Cook et al 2016). Participants consumed a capsule in the morning
193 and in the evening with a meal. On day-7, participants consumed both capsules, 2-hours
194 before the testing. Each NZBC capsule contained 105 mg of anthocyanins, giving a daily
195 intake of 210 mg anthocyanins from the capsules. Random allocation resulted in eleven
196 participants receiving the placebo on visit three (three males and eight females). An exit
197 survey during the visit-four debriefs identified that nine participants (three males and six
198 females) correctly guessed the experimental condition order they received (41%). For the
199 females, visit three and four were 28-days apart to assume testing in the same phase of the
200 menstrual cycle (Bonen et al., 1983), while for the males, a minimum 14-day washout
201 between conditions was used.

202 For visit three and four, participants exercised at an individualised speed to elicit 50% $\dot{V}O_{2max}$
203 with measurement of heart rate (Polar H7, Polar Electro Oy, Kempele, Finland) and expired
204 gases (Cortex Metalyzer 3B, Biophysik GmbH, Leipzig, Germany) with analysis every 5-
205 minutes by averaging the last 60 s of each 5-minute section. Stoichiometric equations were
206 used to calculate carbohydrate and fat oxidation (Jeukendrup and Wallis 2005). Rating of
207 perceived exertion (Borg 6-20) was recorded every 15-minutes. The exercise was followed
208 by participants resting on a massage plinth for 120-minutes with blood pressure measurement
209 immediately and then every 15-minutes. Participants consumed water *ad libitum*.

210 **Incremental Intensity Oxygen-speed Relationship**

211 The incremental treadmill protocol in visit one allowed the relationship between $\dot{V}O_2$ and
212 speed to be established. Starting at 5 km·hr⁻¹ and 1% incline, the protocol increased by 1
213 km·hr⁻¹ every 4-minutes until 11 km·hr⁻¹. Expired gases were averaged for the last 60s of
214 each stage and combined with $\dot{V}O_{2max}$ and $v\dot{V}O_{2max}$, and relationship analysis allowed
215 calculation of the speed required for 50% $\dot{V}O_{2max}$.

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

217 The protocol commenced at 7 km·hr⁻¹ for females and 8 km·hr⁻¹ for males on a 1% incline,
218 increasing by 1 km·hr⁻¹ every minute until voluntary termination. The square wave protocol
219 commenced at 7 km·hr⁻¹ for 2-minutes before increasing to 110% $v\dot{V}O_{2max}$ until voluntary
220 termination. Participants were verbally encouraged throughout and had no real-time
221 feedback. The highest $\dot{V}O_2$ obtained in both protocols was determined from 15-breath
222 averaging, with 18 participants demonstrating a plateau of < 2.1 mL·kg⁻¹·min⁻¹ in $\dot{V}O_2$
223 between the last two speed increments. The square wave protocol confirmed that there was
224 no additional increase in $\dot{V}O_2$ with the additional speed. Only two participants achieved a
225 higher $\dot{V}O_2$ in the verification protocol (step:43.8±8.6, verification:42.1±8.2 mL·kg⁻¹·min⁻¹,
226 P=0.002).

227 **Blood Pressure**

228 For all blood pressure measurements within this study, participants rested supine on a
229 massage plinth with back angle set to 45° without the use of electronic devices or books. The
230 sphygmomanometer cuff (Omron M5-I, Omron Corporation, Kyoto, Japan) was placed
231 around the upper arm approximately 2cm above the brachial artery. The Omron M5-I passes
232 the validation recommendation of the international protocol of the European Society of
233 Hypertension, with a device-observer difference of 0.2±3.6 mmHg for systolic and 0.2±3.9
234 mmHg for diastolic against the mercury sphygmomanometer technique (Omboni et al 2007).

235 Two measurements were taken, and the lowest systolic (SBP) and diastolic (DBP) pressure
236 recorded. Mean arterial pressure (MAP) was calculated by:

$$237 \text{ MAP} = \text{DBP} + [(\text{SBP} - \text{DBP}) \div 3]$$

238 **Statistical analysis and calculations**

239 Data was analysed using SPSS (version 27.0) and GraphPad (version 9.4.1 GraphPad
240 Software). A α -priori power analysis indicated that a sample size of 22 was required to
241 determine a within-between interaction of a 25% increase in FATOX between the placebo
242 and NZBC in the males and females (Willems et al 2022), with a high statistical power
243 (G*Power: $1 - \beta = 0.08$, $\alpha = 0.05$). Data was assessed using Shapiro-Wilk test for normality.
244 Condition (i.e., NZBC vs. PLA vs. no exercise), sex (male vs. female), time, and interaction
245 effects were analysed with a repeated measures ANOVA. Specificity was checked using
246 Mauchly's test, and if violated, Greenhouse-Geisser correction was applied. Post hoc analysis
247 was completed with Bonferroni corrected comparisons. Delta change (Δ) for the 120-minutes
248 average blood pressure changes was also calculated for placebo and NZBC against the no
249 exercise condition. The fat and carbohydrate oxidation following intake of placebo was
250 subtracted (i.e., absolute change) from the oxidation rate with intake of NZBC extract (i.e.,
251 Δ FATOX and Δ CHOOX). Respiratory exchange ratio (RER) was calculated as the ratio
252 between the carbon dioxide produced and oxygen consumed. Cohen's d effect size (Cohen,
253 1998) was calculated with interpretation as <0.2 trivial, $0.2-0.49$ small, $0.5-0.69$ moderate,
254 and >0.8 large. Data is presented as mean \pm standard deviation.

255 **RESULTS**

256 **RPE**

257 For the 1-hour treadmill run at 50% $\dot{V}O_{2max}$, there was a change over time for RPE ($P<0.001$),
258 but no difference between the conditions, or interactions between condition and sex, time and
259 sex or condition and time ($P>0.05$).

260 **Metabolic Responses during Exercise**

261 For the 1-hour treadmill run at 50% $\dot{V}O_{2max}$, participants exercised at 6.8 ± 1.1 km·hr⁻¹ and
262 there was a difference in the speed between males (7.5 ± 1.1 km·hr⁻¹) and females (6.0 ± 0.4
263 km·hr⁻¹, $P<0.001$). However, there was no difference in average relative intensity for NZBC
264 and placebo between males (NZBC: 51.7 ± 6.1 , PLA: 49.5 ± 6.9 % $\dot{V}O_{2max}$) and females (NZBC:
265 52.1 ± 6.9 , PLA: 52.9 ± 9.2 % $\dot{V}O_{2max}$) ($P=0.109$).

266 **For the** absolute $\dot{V}O_2$ there was a change over time ($P=0.036$), but no effect of condition, or
267 interactions ($P>0.05$). The change over time was also observed for relative $\dot{V}O_2$ ($P=0.010$),
268 economy ($P=0.006$), \dot{V}_E ($P<0.001$) with no interactions or condition differences. The $\dot{V}CO_2$
269 demonstrated no time, condition, or sex effects ($P>0.05$). Heart rate changed over time
270 ($P=0.002$) and over time between sex ($P<0.001$) (Table 2).

271 RER demonstrated a change over time ($P<0.001$) and a difference between the conditions
272 ($P<0.001$), with no interactions between condition and sex ($P=0.700$), time and condition
273 ($P=0.102$) and time and sex ($P=0.539$). RER was different (**$P<0.05$**) at 5, 10, 15, 20, 25, 30,
274 35, 50 and 60-minutes of exercise (Figure 1 A).

275 The **FATOX** demonstrated an effect on condition ($P<0.001$), and time ($P<0.001$), but no
276 interactions of condition and sex ($P=0.225$), time and sex ($P=0.349$) or condition and time
277 ($P=0.610$). *Post hoc* comparisons between conditions indicated differences (**$P<0.05$**) at 5
278 ($d=0.588$), 10 ($d=0.577$), 15 ($d=0.518$), 20 ($d=1.134$), 25 ($d=0.542$), 30 ($d=0.932$), 35
279 ($d=0.912$) 50 ($d=0.520$), 55 ($d=0.572$) and 60-minutes ($d=0.588$) (Figure 1D). Average fat
280 oxidation for males and females during the exercise was different between the conditions

281 (NZBC:0.27±0.11, PLA:0.21±0.12 g·min⁻¹, P<0.001, d=0.859), but the responses between
282 males (NZBC:0.27±0.11, PLA:0.19±0.11 g·min⁻¹, P=0.012, d=0.921) and females was not
283 different (P=0.078) (NZBC:0.26±0.12, PLA:0.22±0.14 g·min⁻¹, P=0.014, d=0.899).

284 Similarly, carbohydrate oxidation demonstrated an effect on condition (P=0.002), and time
285 (P<0.001), but no interactions of condition and sex (P=0.192), time and sex (P=0.305) or
286 condition and time (P=0.896). There were differences (P<0.05) at 20 (d=-0.888), 30 (d=-
287 0.477), 40 (d=-0.467), 50 (d=-0.466), 55 (d=-0.456) and 60-minutes (d=-0.536) (Figure 1G).
288 Combined average carbohydrate oxidation for males and females during the exercise was
289 different between the conditions (NZBC:1.32±0.44, PLA:1.43±0.49 g·min⁻¹, P=0.002, d=-
290 0.762), but the responses between males (NZBC:1.64±0.33, PLA:1.80±0.38 g·min⁻¹,
291 P=0.011, d=-0.935) and females was not different (P=0.197) (NZBC:0.99±0.23,
292 PLA:1.06±0.27 g·min⁻¹, P=0.0892).

293 **Fat and Carbohydrate Oxidation Responses to Body Composition**

294 When combined there was no relationship (P>0.05) between body fat percentage, fat-free-
295 mass, and fat-mass for ΔFATOX following NZBC (r=-0.079, r=0.141, r=0.012, respectively)
296 (Figure 2A) and ΔCHOOX (r=0.199, r=-0.044, r=0.232, respectively) (Figure 2B). For males
297 there was also no relationship (P>0.05) between body fat percentage, fat-free-mass, and fat-
298 mass for ΔFATOX (r=-0.069, r=0.039, r=0.029, respectively) (Figure 2A) and ΔCHOOX
299 from NZBC (r=-0.071, r=0.076, r=0.029, respectively) (Figure 2B). However, for females
300 there was a relationship between body fat percentage and ΔFATOX (r=0.691, p=0.019)
301 (Figure 2C) but no relationship (P>0.05) between fat-free-mass and fat-mass for ΔFATOX
302 (r=-0.429, r=0.538, respectively). There was no relationship (P>0.05) between body fat
303 percentage, fat-free-mass, and fat-mass for ΔCHOOX in females from NZBC (r=0.14,
304 r=0.440, r=0.158, respectively) (Figure 2D).

Blood Pressure Responses

305 **Blood Pressure Responses**

306 For the 120-minute blood pressure measurement, SBP demonstrated an effect on time
307 ($P=0.005$), condition ($P<0.001$), and interaction of time and condition ($P=0.002$). There was
308 no interaction of sex and time ($P=0.719$), or condition and sex ($P=0.159$). *Post hoc*
309 comparisons indicated at 15-minutes NZBC was different to placebo ($P=0.004$, $d=0.32$). At
310 30 minutes **no exercise** was different to NZBC ($P=0.005$, $d=0.51$), with NZBC also different
311 to placebo ($P=0.019$, $d=0.25$). At 45-minutes **no exercise** was different to NZBC ($P=0.029$,
312 $d=0.49$). At 60-minutes **no exercise** was different to NZBC ($P<0.001$, $d=0.67$), with NZBC
313 also different to placebo ($P<0.001$, $d=0.37$). At 75-minutes **no exercise** was different to
314 NZBC ($P<0.001$, $d=0.73$) and placebo ($P=0.007$, $d=0.43$). At 90-minutes **no exercise** was
315 different to NZBC ($P<0.001$, $d=0.86$) and placebo ($P=0.003$, $d=0.60$), with NZBC also
316 different to placebo ($P=0.043$, $d=0.22$). At 105-minutes **no exercise** was different to NZBC
317 ($P<0.001$, $d=0.77$) and placebo ($P=0.001$, $d=0.61$). At 120-minutes **no exercise** was different
318 to NZBC ($P<0.001$, $d=0.62$), with NZBC also different to placebo ($P=0.014$, $d=0.35$) (Figure
319 3A).

320 The DBP was different over time ($P=0.003$) but not different between conditions or with any
321 interactions ($P>0.05$) (Figure 3D).

322 The MAP was different over time ($P<0.001$), between the conditions ($P=0.002$) and with an
323 interaction between time and condition ($P=0.017$), with no interaction of condition and sex
324 ($P=0.163$) or time and sex ($P=0.355$). At 45-minutes **no exercise** was different to NZBC
325 ($P=0.031$, $d=0.48$). At 60-minutes **no exercise** was different to NZBC ($P=0.026$, $d=0.50$),
326 with NZBC also different to placebo ($P=0.029$, $d=0.42$). At 75-minutes **no exercise** was
327 different to NZBC ($P<0.001$), with NZBC also different to placebo ($P=0.006$, $d=0.67$). At 90-
328 minutes **no exercise** was different to NZBC ($P=0.006$, $d=0.64$). At 120-minutes **no exercise**

329 was different to NZBC (P=0.025, d=0.58), with NZBC also different to placebo (P=0.019,
330 d=0.43) (Figure 3G).

331 The Δ SBP was larger following NZBC than PLA (no exercise vs. NZBC:-5.5±5.4, no
332 exercise vs. PLA:-2.9±5.1 mmHg, P<0.001, d=-0.817) with no difference in responses
333 between males (P=0.123) and females (P=0.149). There was also the same response for
334 Δ DBP (no exercise vs. NZBC:-2.5±5.4 vs. no exercise vs. PLA:-0.9±6.9 mmHg, P=0.013,
335 d=-0.581), with no difference in responses between the males (P=0.185) and females
336 (P=0.276). As a result, the Δ MAP was also larger following NZBC (no exercise vs. NZBC:-
337 3.5±4.7, vs. no exercise vs. PLA:-1.4±5.4, P=0.011, d=-0.598) with no differences between
338 males (P=0.106) and females (P=0.245).

339 **DISCUSSION**

340 This study observed that blood pressure following 60-minutes treadmill exercise at
341 50% $\dot{V}O_{2max}$ was decreased with NZBC extract and these responses were similar in males and
342 females. During the exercise, NZBC extract increased FATOX for males and females with a
343 large effect, however, only females demonstrated a relationship between body fat percentage
344 and magnitude of change in FATOX.

345 **Substrate utilisation during exercise**

346 The increased FATOX following NZBC extract in the present study supports previous
347 observations (Cook et al., 2015, Cook et al., 2017; Strauss et al., 2018; Şahin et al., 2022;
348 Willems et al., 2022; Shan and Cook 2023). The novelty in the present study was that the
349 presence of a cohort of males and females allowed direct comparison of responses in males
350 and females, with females demonstrating a strong relationship between body fat percentage
351 and magnitude of change of FATOX, but males not demonstrating this relationship.

352 Willems et al. (2022) demonstrated that **FATOX** increases during treadmill walking
353 following NZBC extract in females was correlated to body composition of the trunk.
354 Similarly, Şahin et al. (2022) demonstrated a correlation between body fat percentage and the
355 absolute change in **FATOX** with NZBC extract in males in moderate intensity treadmill
356 walking.

357 The comparison of males and females is important, and this study found no relationship ($R=-$
358 0.079) when analysed together ($n=22$), however, when separate ($n=11$) the relationships
359 between body fat percentage and magnitude of change in females becomes apparent. Factors
360 pertaining to individuals responding or non-responding are important for implementation of a
361 supplement. Previous observations demonstrate variability in **FATOX** to NZBC extract, with
362 females demonstrating higher responses. For example, Willems et al. (2022) observed that
363 increased **FATOX** by NZBC extract in untrained physically active females ranged from 10-
364 66%, while Cook et al. (2017) observed in male cyclists responses from 0-44% (unpublished
365 analysis). **The mechanisms for the findings in the present study are not known but may relate**
366 **to fat transport and metabolism. For example**, females have greater FAT/CD36 in comparison
367 to males at all levels of training (Kiens et al., 2004), and higher ratio of maximal **FATOX** rate
368 to fat free mass (Randell et al., 2017). Therefore, these differences may explain the results
369 observed in the present study. However, **it should be recognised that a limitation was that**
370 **there was no control for menstrual cycle status, phase, and use of contraception and this**
371 **should be implemented in future investigations.**

372 **Blood pressure responses**

373 The larger decrease in post exercise blood pressure with NZBC extract repeats a previous
374 observation (Shan and Cook 2023), with a similar Δ SBP of -5.5 mmHg. However, to the

375 authors knowledge, this is the first study to compare responses between males and females,
376 with no differences observed.

377 A consideration of this study is that blood pressure measurements were taken for 120-minutes
378 following exercise under laboratory conditions in participants without hypertension Post-
379 exercise blood pressure decrements can last for 24 hours in hypertensive males and females
380 (Wallace et al., 1997), and it is not known if the effects of NZBC extract on post-exercise
381 blood pressure also lasts for this duration, or in free-living conditions. Furthermore, blood
382 pressure reductions following physical activity are greatest in those with higher resting blood
383 pressure (Pescatello et al., 2018). Therefore, it is also unclear whether NZBC extract induced
384 post-exercise decrements would also work in those who are pre or hypertensive and should be
385 examined in the future. Furthermore, there was no dietary control used within this study
386 except caffeine and alcohol. Micronutrients such as sodium (Huang et al. 2020) are known to
387 influence blood pressure, therefore, future studies must control this.

388 Similarly, to Shan and Cook (2023), the present study measured resting blood pressure in
389 visit two for 120-minutes with no exercise. Therefore, the SBP differences between the
390 placebo and no exercise condition at 75, 90 and 105 minutes (Figure 3A) demonstrate the
391 exercise duration and intensity was sufficient to generate a decrease in blood pressure. The
392 NZBC extract generated a greater decrease in blood pressure above the placebo condition
393 with differences at 15, 30, 60, 90 and 120 minutes. However, the mechanisms are unclear and
394 may be caused by anthocyanin induced increase in nitric oxide availability and arterial
395 vasodilation. For example, *in-vitro* observations demonstrate cyanidin-3-glucoside to increase
396 the gene expression of nitric oxide synthase (Xu et al., 2004) and enter vascular endothelia
397 smooth cells (Zibera et al., 2012). However, to the authors knowledge, there are no *in-vivo*
398 studies in humans that demonstrate a causal link plasma anthocyanins or metabolites and
399 arterial vasodilation. It should also be recognised that this study was powered to examine the

400 metabolic changes between males and females and may be underpowered to detect sex
401 differences in post exercise blood-pressure.

402 **Conclusions**

403 A 7-day intake of NZBC extract increased **FATOX** during 60-minutes of moderate intensity
404 treadmill exercise in males and females, with the magnitude of responses correlated to body
405 fat percentage in females but not in males. In the 120-minutes post-exercise, NZBC extract
406 caused a larger blood pressure decrement than placebo and this response was **not different**
407 **between** males and females.

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412 University of Worcester, United Kingdom.

413 **Data availability**

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

415

416 **Author Contributions:**

417 Conceived and planned the experiments: [REDACTED]

418 Carried out the experiments: [REDACTED]

419 Undertook data analysis: [REDACTED]

420 Wrote the manuscript: [REDACTED]

421

422 **References**

- 423 1. Al-Nakkash, L., Martin, J.B., Petty, D., Lynch, S.M., Hamrick, C., Lucy, D.,
424 Robinson, J., Peterson, A., Rubin, L.J., Broderick, T.L. (2012). Dietary genistein
425 induces sex-dependent effects on murine body weight, serum profiles, and vascular
426 function of thoracic aortae. *Gender Medicine*, 9(5), 295–308.
- 427 2. Bonen, A., Haynes, F.J., Watson-Wright, W., Sopper, M.M., Pierce, G.N., Low, M.P.,
428 Graham, T.E. (1983). Effects of menstrual cycle on metabolic responses to exercise.
429 *Journal of Applied Physiology: Respiratory, Environmental and Exercise Physiology*,
430 55, 1506–1513.
- 431 3. Braakhuis, A.J., Somerville, V.X., & Hurst, R.D. (2020). The effect of New Zealand
432 blackcurrant on sport performance and related biomarkers: A systematic review and
433 meta-analysis [Erratum in: *Journal of the International Society of Sports Nutrition*,
434 (2021);18(1), 8]. *Journal of the International Society of Sports Nutrition*, 17(1), 25.
435 <https://doi.org/10.1186/s12970-020-00354-9>
- 436 4. Burton-Freeman, B., Linares, A., Hyson, D., Kappagoda, T. (2010). Strawberry
437 modulates LDL oxidation and postprandial lipemia in response to high-fat meal in
438 overweight hyperlipidemic men and women. *Journal of the American College of*
439 *Nutrition*, 29, 46–54.
- 440 5. Campesi, I., Marino, M., Cipolletti, M., Romani, A., Franconi, F. (2018). Put “gender
441 glasses” on the effects of phenolic compounds on cardiovascular function and
442 diseases. *European Journal of Nutrition*, 57, 2677–2691.
- 443 6. Cappellini, F., Marinelli, A., Toccaceli, M., Tonelli, C., Petroni, K (2021).
444 Anthocyanins: from mechanisms of regulation in plants to health benefits in foods.
445 *Frontiers in Plant Science*, 12, 748049. doi: 10.3389/fpls.2021.748049

- 446 7. Cerletti, C., De Curtis, A., Bracone, F., Digesi, C., Morganti, A.G., Iacoviello, L., de
447 Gaetano, G., Donati, M.B (2017). Dietary anthocyanins and health: data from FLORA
448 and ATHENA EU projects. *British Journal of Clinical Pharmacology*, 83, 103–106.
449 doi: 10.1111/bcp.12943
- 450 8. Cohen, J. (1998). *Statistical power analysis for the behavioral sciences* (2nd ed.).
451 Erlbaum Associates.
- 452 9. Cook, M.D., & Willems, M.E.T. (2019). Dietary anthocyanins: A review of the
453 exercise performance effects and related physiological responses. *International*
454 *Journal of Sports Nutrition and Exercise Metabolism*, 29(3), 322–330.
455 <https://doi.org/10.1123/ijsnem.2018-0088>
- 456 10. Cook, M.D., Myers, S.D., Blacker, S.D., & Willems, M.E. (2015). New Zealand
457 blackcurrant extract improves cycling performance and fat oxidation in cyclists.
458 *European Journal of Applied Physiology*, 115(11), 2357–2365.
459 <https://doi.org/10.1007/s00421-015-3215-8>
- 460 11. Cook, M.D., Myers, S.D., Gault, M.L., & Willems, M.E.T. (2017). Blackcurrant alters
461 physiological responses and femoral artery diameter during sustained isometric
462 contraction. *Nutrients*, 9(6), 556. <https://doi.org/10.3390/nu9060556>
- 463 12. Cook, M.D., Myers, S.D., Gault, M.L., Edwards, V.C., Willems, M.E. (2017)
464 Cardiovascular function during supine rest in endurance-trained males with New
465 Zealand blackcurrant: a dose-response study. *European Journal of Applied*
466 *Physiology*, 117(2):247-254. <https://doi.org/10.1007/s00421-016-3512-x>.
- 467 13. Elliott-Sale, K.J., Minahan, C.L., de Jonge, X.A.K.J., Ackerman, K.E., Sipilä, S.,
468 Constantini, N.W., Lebrun, C.M., Hackney, A.C. (2021). Methodological
469 Considerations for Studies in Sport and Exercise Science with Women as Participants:

- 470 A Working Guide for Standards of Practice for Research on Women. *Sports*
471 *Medicine*, 51(5), 843861. <https://doi.org/10.1007/s40279-021-01435-8>.
- 472 14. Frandsen, J., Vest, S.D., Larsen, S., Dela, F., Helge, J.W (2017). Maximal Fat
473 Oxidation is Related to Performance in an Ironman Triathlon. *International Journal of*
474 *Sports Medicine*, 38(13), 975-982. <https://doi.org/10.1055/s-0043-117178>
- 475 15. Goodpaster, B.H., Wolfe, R.R., Kelley, D.E. (2002) Effects of obesity on substrate
476 utilization during exercise. *Obesity Research*, 10(7):575-84.
477 <https://doi.org/10.1038/oby.2002.78>.
- 478 16. Guo, J., Han, W., Wang, M. (2008). Ultraviolet and environmental stresses involved
479 in the induction and regulation of anthocyanin biosynthesis: A review. *African*
480 *Journal of Biotechnology*, 7(25), 6.
- 481 17. Huang, L., Trieu, K., Yoshimura, S., Neal, B., Woodward, M., Campbell, N.R.C., Li,
482 Q., Lackland, D.T., Leung, A.A., Anderson, C.A.M., MacGregor, G.A., He, F.J.
483 (2020). Effect of dose and duration of reduction in dietary sodium on blood pressure
484 levels: systematic review and meta-analysis of randomised trials. *BMJ*. 24;368:m315.
- 485 18. Jeukendrup, A.E., & Wallis, G.A. (2005). Measurement of substrate oxidation during
486 exercise by means of gas exchange measurements. *International Journal of Sports*
487 *Medicine*, 26(Suppl. 1), S28–S37. <https://doi.org/10.1055/s-2004-830512>
- 488 19. Keane, K.M., George, T.W., Constantinou, C.L., Brown, M.A., Clifford, T.,
489 Howatson, G. (2016). Effects of Montmorency tart cherry (*Prunus Cerasus L.*)
490 consumption on vascular function in men with early hypertension. *American Journal*
491 *of Clinical Nutrition*, 103(6), 1531–1539. <https://doi.org/10.3945/ajcn.115.123869>
- 492 20. Kiens, B., Roepstorff, C., Glatz, J.F., Bonen, A., Schjerling, P., Knudsen, J., Nielsen,
493 J.N. (2004). Lipid-binding proteins and lipoprotein lipase activity in human skeletal

- 494 muscle: influence of physical activity and gender. *Journal of Applied Physiology*,
495 97(4),1209-1218. <https://doi.10.1152/jappphysiol.01278.2003>.
- 496 21. Omboni, S., Riva, I., Giglio, A., Caldara, G., Groppelli, A., Parati, G. (2007).
497 Validation of the Omron M5-I, R5-I and HEM-907 automated blood pressure
498 monitors in elderly individuals according to the International Protocol of the
499 European Society of Hypertension. *Blood Pressure Monitoring*, 12(4):233-42.
500 <https://doi.org10.1097/MBP.0b013e32813fa386>.
- 501 22. Pescatello, L.S., Buchner, D.M., Jakicic, J.M., Powell, K.E., Kraus, W.E., Bloodgood,
502 B., Campbell, W.W., Dietz, S., Dipietro, L., George, S.M., Macko, R.F., McTiernan,
503 A., Pate, R.R., Piercy, K.L. (2018). Physical Activity to Prevent and Treat
504 Hypertension: A Systematic Review. *Medicine & Science in Sports & Exercise*, 51(6),
505 1314-1323. <https://doi.org10.1249/MSS.0000000000001943>
- 506 23. Poole, D.C., & Jones, A.M. (2017). Measurement of the maximum oxygen uptake
507 VO₂max: VO₂peak is no longer acceptable. *Journal of Applied Physiology*, 122(4),
508 997–1002. <https://doi.org/10.1152/jappphysiol.01063.2016>
- 509 24. Queiroz, A.C., Rezk, C.C., Teixeira, L., Tinucci, T., Mion, D., Forjaz, C.L. (2013)
510 Gender influence on post-resistance exercise hypotension and hemodynamics.
511 *International Journal of Sports Medicine*, 34(11):939-44. [https://doi.org/10.1055/s-0033-](https://doi.org/10.1055/s-0033-1337948)
512 [1337948](https://doi.org/10.1055/s-0033-1337948).
- 513 25. Randell, R.K., Rollo, I., Roberts, T.J., Dalrymple, K.J., Jeukendrup, A.E., Carter, J.M
514 (2017). Maximal Fat Oxidation Rates in an Athletic Population. *Medicine & Science*
515 *in Sports & Exercise*, 49(1),133-140.
516 <https://doi.org.10.1249/MSS.0000000000001084>.
- 517 26. Rothwell, J.A., Perez-Jimenez, J., Neveu, V., Medina-Remón, A., M'hiri, N., García-
518 Lobato, P., Manach, C., Knox, C., Eisner, R., Wishart, D.S., Scalbert, A. (2013)

- 519 Phenol-Explorer 3.0: a major update of the Phenol-Explorer database to incorporate
520 data on the effects of food processing on polyphenol content. *Database (Oxford)*,
521 <https://doi.org/10.1093/database/bat070>.
- 522 27. Şahin, M.A., Bilgiç, P., Montanari, S., & Willems, M.E.T. (2022). Daily and not
523 every-other-day intake of anthocyanin-rich New Zealand blackcurrant extract alters
524 substrate oxidation during moderate-intensity walking in adult males. *Journal of*
525 *Dietary Supplements*, 19(1), 49–61. <https://doi.org/10.1080/19390211.2020.1841356>
- 526 28. Shan Y, Cook MD (2023). New Zealand Blackcurrant Increases Postexercise
527 Hypotension Following Sustained Moderate-Intensity Exercise. *International Journal*
528 *of Sport Nutrition and Exercise Metabolism*, 33(5), 282-290.
529 <https://doi.org/10.1123/ijsnem.2023-0014>.
- 530 29. Smith, E.S., McKay, A.K.A., Kuikman, M., Ackerman, K.E., Harris, R., Elliott-Sale,
531 K.J., Stellingwerff, T., Burke, L.M. (2022). Auditing the Representation of Female
532 Versus Male Athletes in Sports Science and Sports Medicine Research: Evidence-
533 Based Performance Supplements. *Nutrients*, 14(5), 953.
534 <https://doi.org/10.3390/nu14050953>
- 535 30. Strauss, J.A., Willems, M.E.T., & Shepherd, S.O (2018). New Zealand blackcurrant
536 extract enhances fat oxidation during prolonged cycling in endurance-trained females.
537 *European Journal of Applied Physiology*, 118(6), 1265–1272.
538 <https://doi.org/10.1007/s00421-018-3858-3>
- 539 31. Vest, S.D., Frandsen, J., Larsen, S., Dela, F., Helge, J.W (2018). Peak Fat Oxidation
540 is not Independently Related to Ironman Performance in Women. *International*
541 *Journal of Sports Medicine*, 39(12), 916-923. <https://doi.org/10.1055/a-0660-0031>
- 542 32. Wallace, J.P., Bogle, P.G., King, B.A., Krasnoff, J.B., Jastremski, C.A. (1997). A
543 comparison of 24-h average blood pressures and blood pressure load following

- 544 exercise. *American Journal of Hypertension*, 10(7), 728-734.
- 545 [https://doi.org.10.1016/s0895-7061\(97\)00070-8](https://doi.org.10.1016/s0895-7061(97)00070-8).
- 546 33. Willems, M.E.T., & Blacker, S.D. (2022) Anthocyanin-rich supplementation:
547 Emerging evidence of strong potential for sport and exercise nutrition. *Frontiers in*
548 *Nutrition*, 9, 864323. <https://doi.org/10.3389/fnut.2022.864323>
- 549 34. Willems, M.E.T., Banic, M., Cadden, R., & Barnett, L. (2022). Enhanced walking-
550 induced fat oxidation by New Zealand blackcurrant extract is body composition-
551 dependent in recreationally active adult females. *Nutrients*, 14(7), 1475.
552 <https://doi.org/10.3390/nu14071475>
- 553 35. Willems, M.E.T., Myers, S.D., Gault, M.L., & Cook, M.D. (2015). Beneficial
554 physiological effects with blackcurrant intake in endurance athletes. *International*
555 *Journal of Sport Nutrition and Exercise Metabolism*, 25(4), 367–374.
556 <https://doi.org/10.1123/ijsnem.2014-0233>
- 557 36. Williams, B., Mancia, G., Spiering, W., Agabiti, Rosei E., Azizi, M., Burnier, M.,
558 Clement, D.L., Coca, A., de Simone, G., Dominiczak, A., Kahan, T., Mahfoud, F.,
559 Redon, J., Ruilope, L., Zanchetti, A., Kerins, M., Kjeldsen, S.E., Kreutz, R., Laurent,
560 S., Lip, G.Y.H., McManus, R., Narkiewicz, K., Ruschitzka, F., Schmieder, R.E.,
561 Shlyakhto, E., Tsioufis, C., Aboyans, V., Desormais, I. (2018). 2018 ESC/ESH
562 Guidelines for the management of arterial hypertension: The Task Force for the
563 management of arterial hypertension of the European Society of Cardiology and the
564 European Society of Hypertension: The Task Force for the management of arterial
565 hypertension of the European Society of Cardiology and the European Society of
566 Hypertension. *Journal of Hypertension*, 36(10):1953-2041.
567 [doi:10.1097/HJH.0000000000001940](https://doi.org/10.1097/HJH.0000000000001940).

- 568 37. World Medical Association. (2013). World Medical Association Declaration of
569 Helsinki: Ethical principles for medical research involving human subjects. *JAMA*,
570 *310*(20), 2191–2194. <https://doi.org/10.1001/jama.2013.281053>
- 571 38. Xu, J.W., Ikeda, K., & Yamori, Y. (2004). Cyanidin-3-glucoside regulates
572 phosphorylation of endothelial nitric oxide synthase. *FEBS Letters*, *574*(1–3), 176–
573 180. <https://doi.org/10.1016/j.febslet.2004.08.027>
- 574 39. Yi, L., Chen, C.Y., Jin, X., Mi, M.T., Yu, B., Chang, H., Ling, W.H., Zhang, T.
575 (2010). Structural requirements of anthocyanins in relation to inhibition of endothelial
576 injury induced by oxidized low-density lipoprotein and correlation with radical
577 scavenging activity. *FEBS Letters*, *5*;584(3):583-90.
578 <https://doi.org:10.1016/j.febslet.2009.12.006>.
- 579 40. Ziberna, L., Tramer, F., Moze, S., Vrhovsek, U., Mattivi, F., & Passamonti, S. (2012).
580 Transport and bioactivity of cyanidin 3-glucoside into the vascular endothelium. *Free*
581 *Radical Biology and Medicine*, *52*(9), 1750–1759.
582 <https://doi.org/10.1016/j.freeradbiomed.2012.02.027>
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591 Table 1 Male and female participant characteristics

	Male (n=11)	Female (n=11)	Combined (n=22)	P
Age (years)	32±9	27±7	29±8	0.172
Height (cm)	178±7	163±6*	171±10	<0.001
Body Mass (kg)	73±13	64±7	69±11	0.060
Body Mass Index	23.0±3.4	24.2±2.1	23.6±2.9	0.331
Body Fat (%)	13.1±2.2	23.4±4.7*	18±6	<0.001
Fat Free Mass (kg)	64±12	49±5*	56±12	<0.001
Fat Mass (kg)	10±2	15±4*	13±4	<0.001
$\dot{V}O_{2max}$ (mL·kg ⁻¹ ·min ⁻¹)	50±6	38±7*	44±9	<0.001
$\dot{V}O_{2max}$ (L·min ⁻¹)	3.6±0.5	2.5±0.6*	3.0±0.8	<0.001
$v\dot{V}O_{2max}$ (km·hr ⁻¹)	18.5±2.2	14.6±1.7*	16.6±2.8	<0.001
RER _{max} (AU)	1.18±0.08	1.20±0.03	1.19±0.07	0.348
HR _{max} (b·min ⁻¹)	185±12	191±13	188±12	0.313
Resting Systolic Pressure (mmHg)	120±9	113±11	116±11	0.128
Resting Diastolic Pressure (mmHg)	71±8	72±6	72±7	0.806
Resting Mean Arterial Pressure (mmHg)	87±7	86±8	86±7	0.573
Resting Rate Pressure Product	7055±1162	6755±1109	6605±1808	0.541
Classification of Blood Pressure				
Optimal (Sys<120 and dia<80)	7	9	16	-
Normal (Sys 120-129 and/or dia 80-84)	2	2	4	-
High Normal (Sys 130-139 and/or dia 85-89)	2	0	2	-

592 Data reported as mean±SD and counts. $\dot{V}O_{2max}$ = maximal oxygen update; $v\dot{V}O_{2max}$ = running speed at $\dot{V}O_{2max}$;

593 RER_{max} = maximum respiratory exchange ratio; HR_{max} = maximum heart rate; AU = arbitrary units. *females

594 different to males (P<0.05).

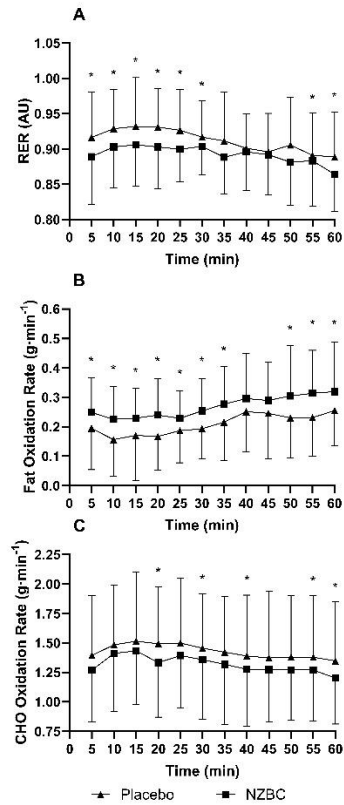
595 Table 2 Volume of oxygen uptake and carbon dioxide produced, heart rate, economy, minute ventilation during 60-minutes of treadmill exercise
 596 following placebo and NZBC extract in males and females

	Time (min)												
	5	10	15	20	25	30	35	40	45	50	55	60	
$\dot{V}CO_2$ (L·min⁻¹)													
Combined NZBC	1.41±0.40	1.42±0.41	1.44±0.41	1.42±0.40	1.44±0.41	1.43±0.40	1.42±0.41	1.41±0.41	1.40±0.41	1.41±0.39	1.39±0.39	1.39±0.38	
Combined PLA	1.37±0.38	1.40±0.36	1.43±0.37	1.40±0.36	1.39±0.39	1.40±0.38	1.40±0.39	1.40±0.38	1.40±0.37	1.38±0.35	1.39±0.37	1.38±0.35	
Male NZBC	1.71±0.26	1.73±0.31	1.74±0.32	1.71±0.31	1.72±0.35	1.71±0.33	1.72±0.32	1.72±0.30	1.70±0.31	1.69±0.31	1.67±0.32	1.67±0.28	
Male PLA	1.60±0.35	1.64±0.29	1.71±0.24	1.63±0.31	1.63±0.34	1.64±0.33	1.65±0.36	1.65±0.33	1.66±0.26	1.63±0.28	1.67±0.24	1.65±0.17	
Female NZBC	1.10±0.25	1.12±0.24	1.14±0.23	1.12±0.24	1.16±0.23	1.14±0.22	1.11±0.22	1.10±0.22	1.10±0.23	1.12±0.21	1.11±0.21	1.10±0.23	
Female PLA	1.13±0.24	1.17±0.24	1.15±0.24	1.17±0.24	1.14±0.26	1.16±0.25	1.15±0.23	1.15±0.24	1.13±0.25	1.14±0.22	1.12±0.24	1.11±0.26	
$\dot{V}O_2$ (mL·kg⁻¹·min⁻¹)[†]													
Combined NZBC	22.6±4.8	22.6±5.0	22.7±4.7	22.5±4.7	22.9±5.0	23.0±5.1	23.0±5.0	22.9±5.1	22.7±5.0	23.0±4.8	22.8±4.7	22.7±4.5	
Combined PLA	21.9±4.8	22.0±4.4	22.5±4.6	22.0±4.6	21.9±4.7	22.3±4.8	22.6±5.0	22.6±4.9	22.7±4.6	22.4±4.5	22.7±4.6	22.8±4.7	
Male NZBC	25.6±3.7	25.7±4.3	25.5±4.2	25.4±4.0	25.7±4.8	26.1±5.0	26.1±4.6	26.1±4.6	25.7±4.5	25.7±4.5	25.5±4.5	25.5±3.9	
Male PLA	23.8±5.3	23.9±4.5	24.9±4.1	24.0±4.8	24.1±4.6	24.5±4.9	24.7±5.4	24.8±4.9	25.1±4.1	24.6±4.4	25.3±3.9	25.6±3.7	
Female NZBC	19.6±3.8	19.5±3.6	19.9±3.4	19.7±3.5	20.1±3.4	19.9±3.1	20.0±3.4	19.7±3.4	19.7±3.6	20.2±3.3	20.2±3.3	20.0±3.3	
Female PLA	20.0±3.7	20.1±3.7	20.1±3.7	20.0±3.6	19.7±3.6	20.0±3.6	20.4±3.7	20.5±3.9	20.4±4.0	20.2±3.5	20.1±3.7	20.0±4.0	
$\dot{V}O_2$ (L·min⁻¹)[†]													
Combined NZBC	1.55±0.42	1.55±0.43	1.56±0.41	1.55±0.41	1.57±0.43	1.57±0.43	1.58±0.43	1.58±0.44	1.57±0.44	1.58±0.42	1.57±0.41	1.57±0.42	
Combined PLA	1.50±0.39	1.51±0.36	1.54±0.37	1.51±0.38	1.51±0.40	1.53±0.39	1.55±0.41	1.55±0.40	1.56±0.37	1.53±0.37	1.56±0.38	1.56±0.38	
Male NZBC	1.84±0.30	1.85±0.33	1.84±0.33	1.83±0.31	1.85±0.37	1.86±0.36	1.88±0.35	1.88±0.35	1.86±0.36	1.85±0.35	1.84±0.36	1.84±0.34	
Male PLA	1.71±0.37	1.72±0.31	1.79±0.28	1.73±0.34	1.75±0.35	1.76±0.36	1.78±0.39	1.79±0.37	1.80±0.29	1.77±0.31	1.81±0.27	1.83±0.22	
Female NZBC	1.27±0.30	1.26±0.29	1.28±0.27	1.27±0.29	1.29±0.27	1.28±0.27	1.29±0.28	1.28±0.29	1.27±0.30	1.30±0.27	1.30±0.27	1.29±0.29	
Female PLA	1.29±0.29	1.29±0.28	1.29±0.28	1.29±0.28	1.27±0.29	1.29±0.28	1.31±0.28	1.32±0.29	1.32±0.29	1.30±0.27	1.30±0.28	1.29±0.30	
Economy (mL·kg⁻¹·km⁻¹)[†]													
Combined NZBC	199±28	199±26	200±25	198±25	201±25	202±25	202±26	202±28	200±27	203±26	201±26	201±25	
Combined PLA	191±28	193±25	198±26	193±27	192±26	195±27	198±29	199±28	200±28	197±25	200±27	201±31	
Male NZBC	202±20	203±20	203±20	201±20	203±23	206±25	206±22	208±26	203±22	205±25	202±24	203±21	
Male PLA	185±26	187±17	197±22	188±24	190±22	192±24	193±28	195±24	199±22	194±22	201±26	204±30	
Female NZBC	195±34	194±32	197±29	196±30	200±29	198±27	199±29	196±29	196±32	201±28	201±29	198±28	
Female PLA	198±30	199±30	199±31	199±30	195±30	199±30	202±30	203±33	202±34	200±28	200±30	198±34	
Heart Rate (beats·min⁻¹)^{†,#}													
Combined NZBC	124±12	125±12	124±12	126±13	127±15	128±14	130±16	131±17	131±17	132±17	133±17	132±18	
Combined PLA	124±15	126±14	128±14	127±14	127±13	128±13	129±13	129±13	131±14	132±15	133±15	132±16	
Male NZBC	123±16	124±16	124±16	128±18	130±20	131±19	134±21	135±23	136±22	137±23	137±23	137±23	
Male PLA	123±18	125±17	130±19	129±19	130±18	130±19	131±19	132±18	134±19	135±20	136±20	137±21	
Female NZBC	125±10	125±8	124±7	123±8	124±9	125±8	127±9	126±9	127±12	128±8	129±9	127±10	
Female PLA	126±14	127±11	126±8	125±8	125±8	126±7	127±5	127±7	128±6	129±7	129±8	128±9	
\dot{V}_E (L·min⁻¹)[†]													
Combined NZBC	41.9±9.4	44.2±10.6	44.5±9.6	44.5±10.3	46.1±10.9	46.3±12.0	46.2±11.3	46.2±11.6	45.9±11.5	46.6±10.8	46.7±11.0	46.8±11.2	
Combined PLA	42.3±9.7	44.0±9.1	45.2±9.3	44.9±9.1	45.2±10.8	45.4±10.3	45.8±10.5	46.1±10.0	46.0±10.3	46.1±9.8	46.4±10.3	47.1±10.7	
Male NZBC	47.1±9.4	49.6±11.2	49.6±9.5	49.6±11.1	51.5±12.0	52.0±13.4	52.1±12.5	52.4±12.6	51.8±12.6	51.8±11.7	52.3±12.0	52.8±10.4	
Male PLA	47.8±10.5	48.6±9.8	51.0±8.9	49.4±10.0	50.7±11.2	50.9±10.9	51.1±11.3	51.4±10.1	51.6±9.4	51.0±10.3	52.1±9.6	53.3±8.2	
Female NZBC	36.7±5.9	38.8±6.9	39.4±6.8	39.4±6.6	40.6±6.4	40.5±7.1	40.4±6.1	39.9±6.4	40.1±6.7	41.4±6.8	41.2±6.6	40.8±8.7	
Female PLA	36.8±4.6	39.3±5.5	39.5±5.4	40.3±5.3	39.7±7.2	39.9±6.2	40.5±6.3	40.7±6.7	40.4±8.0	41.2±6.6	40.8±7.8	40.8±9.3	

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597 Note. All measures collected following 7-days of supplementation with NZBC extract or placebo during treadmill exercise. Data reported as mean±sd from 22 participants (11 female, 11 male).
598 †significant effect for time (P<0.05), #significant interaction between time and sex (P<0.05).
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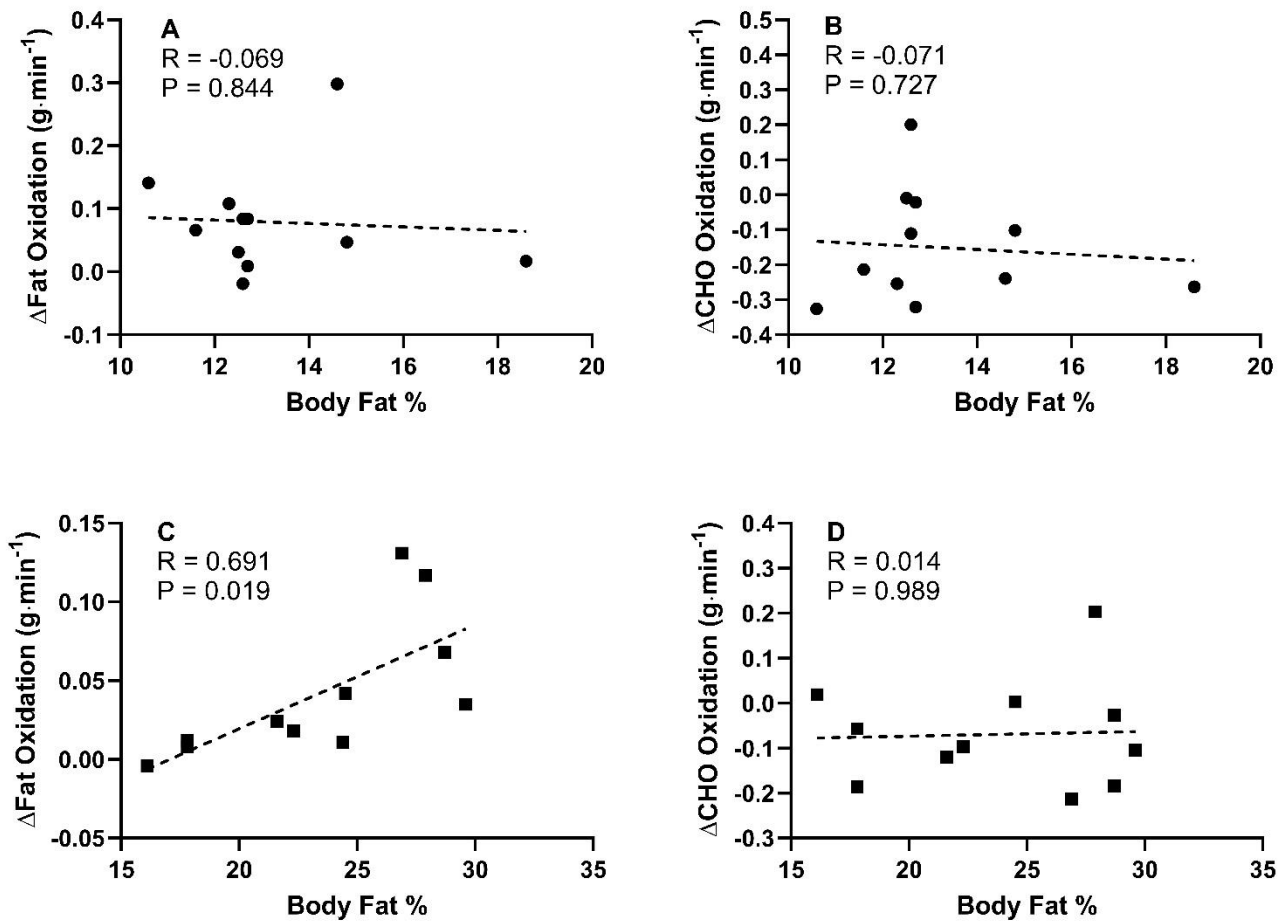
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627 Figure 1 (a) combined RER (b) male RER, (c) female RER, (d) combined fat oxidation, (e) male fat oxidation, (f) female fat oxidation, (g)

628 combined carbohydrate oxidation, (h) male carbohydrate oxidation, (i) female carbohydrate oxidation following 7-days NZBC extract or placebo

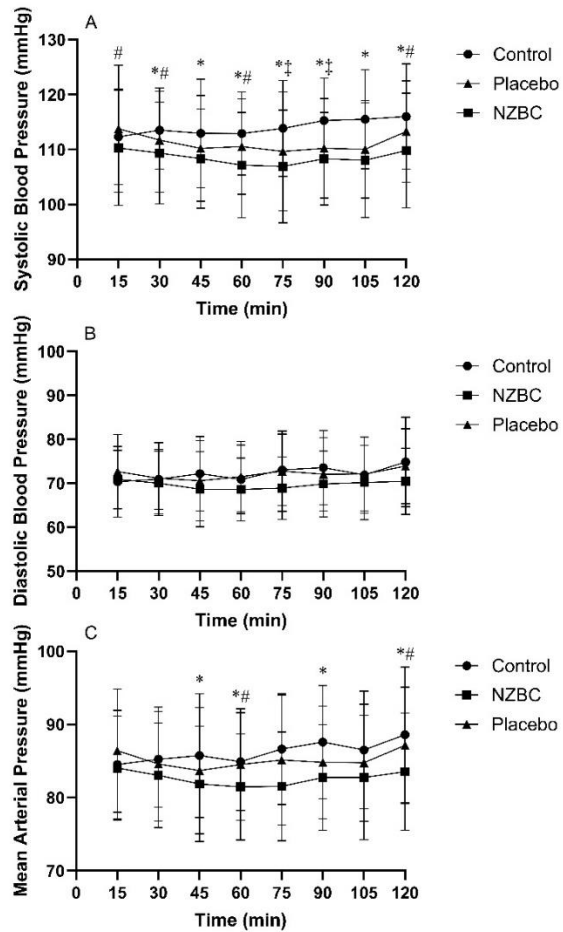
629 during 60-minutes of treadmill exercise at 50% $\dot{V}O_{2max}$ in males and females (n=22 - 11 males and 11 females). Data presented as mean±SD.

630 *NZBC extract different (P<0.05) to placebo.

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 632 Figure 2 (a) male Δ FATOX, (b) male Δ CHOOX, (c) female Δ FATOX, (d) female FOA Δ CHOOX responses following NZBC extract against
 633 body fat percentage.

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635
 636 Figure 3 (a) combined systolic, (b) male systolic, (c) female systolic, (d) combined diastolic, (e) male diastolic, (f) female diastolic, (g)
 637 combined mean arterial pressure, (h) male mean arterial pressure, (i) female mean arterial pressure in the no exercise condition and 120-minutes
 638 following exercise with NZBC extract or placebo. *no exercise different to NZBC extract; ‡ no exercise different to placebo; # NZBC extract
 639 different to placebo.
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