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Blackcurrant extract does not affect the speed-duration relationship during high-intensity running

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ABSTRACT

Anthocyanin-rich blackcurrant extract (BC) has been shown to ergogenically aid high-intensity exercise. Capacity for such exercise is evaluated by the hyperbolic speed-tolerable duration (S-Dtol) relationship. Therefore, in double-blinded and cross-over randomized controlled trials, 15 males underwent treadmill running incremental exercise testing and were assessed for S-Dtol, quantified by critical speed (CS) and D’ (distance), and assessments of time to exhaustion performance to empirically test the limits of the S-Dtol relationship, after daily supplementation of 300mg/d BC (105mg/d anthocyanin) or placebo. Supplementation with BC did not change CS (placebo 12.1±1.0km/h vs BC 11.9±1.0km/h, p>0.05) or D’ (placebo 918.6±223.2m vs BC 965.2±231.2m, p>0.05), although further analysis indicated D’ increased in 60% of subject (p=0.08), indicating a trend toward cohorts potentially benefiting from BC supplementation. BC supplementation did not change time to exhaustion at or above CS, maximal oxygen uptake (VO2max), lactate threshold (LT), submaximal running economy (C_R), or substrate utilization during exercise (all p>0.05). In conclusion, we could not detect any beneficial effect of BC supplementation during high-intensity running exercise, including the determining factors S-Dtol relationship, VO2max, LT or C_R. Hence, no ergogenic effect was observed.

Key words: Anthocyanin; Blackcurrant extract; High-intensity exercise; Critical speed; Speed-tolerable duration.
**Introduction**

Anthocyanins, of the flavonoid group of polyphenols, occur naturally in blackcurrant (Ribes Nigrum, BC) and its concentrated extract. This BC extract has emerged as a commercially available and legal sports supplement with purported ergogenic effects. A full and complete understanding of the phenomenon has not yet been fully established, but the observed effects relate to both health and exercise performance.\(^1\)-\(^3\)

Several mechanisms of action have been considered. The strongest evidence suggest that anthocyanins 1) provide anti-oxidant actions that reduce oxidative stress;\(^2\)-\(^4\) 2) protect the endothelium, promote nitric oxide bioavailability and facilitate endothelial-dependent vasorelaxation;\(^5\)-\(^7\) 3) increase the protective immune responsiveness to potential pathogens;\(^8\),\(^9\) and 4) increase tissue fat oxidation.\(^10\),\(^11\) Thus, these actions have several health implications as they first and foremost protect against conditions exaggerated by oxidative stress and endothelial dysfunction such as cardiovascular disease, diabetes, certain cancers and degenerative diseases.\(^3\)-\(^7\),\(^12\) Secondly, they reduce inflammations,\(^9\) and thirdly, they help manage metabolic problems and overweight.\(^10\),\(^11\) Further studies have indicated that BC and anthocyanin supplementation also may confer broad-spectrum positive acid/base effects,\(^13\) anti-bacterial and anti-viral effects,\(^14\),\(^15\) certain regenerative effects,\(^16\) while also reducing exercise-induced cell and tissue damage,\(^8\),\(^9\),\(^17\) tumor development and proliferation\(^18\) as well as pain, fatigue and morbidity.\(^19\),\(^20\) However, it is not always clear whether the observed action was caused by anthocyanins and their conjugates, other anthocyanin-derived metabolites, or other compounds or derivatives from the anthocyanin source.\(^1\),\(^2\)

The present study was however based on the notion that supplementation of anthocyanin-rich BC extract may also improve exercise performance. This is because, following from the abovementioned mechanisms, anthocyanins in effect may also increase skeletal muscle blood flow and microperfusion, increase blood lactate clearance, and improve skeletal muscle metabolic, contractile and recovery status,\(^1\),\(^3\),\(^8\),\(^11\),\(^17\),\(^21\)-\(^24\) and thereby provide substrate to enhance exercise capacity and recovery. As such, anthocyanin-rich BC supplementation has been evidenced to increase exercise performance, at least under certain conditions. In a series of studies, Willems and colleagues showed that supplementation of mainly 300mg/day New Zealand Blackcurrant Extract (105mg/day anthocyanins) improved 1) 16.1km (~28-29mins) cycling time-trial performance by 2.4% (44secs);\(^10\) 2) high-intensity intermittent cycling time trial performance, measured as total time to completion, by ~1% (3-4secs in each 4km
3) high-intensity intermittent sprint running performance, measured as individual and accumulated sprint distances, by 10-11%;\textsuperscript{22} and 4) high-intensity intermittent shuttle running sprint performance, measured as reduced decrement in sprint time from first to last sprint by 40-50%,\textsuperscript{26,27} compared to placebo.

In contrast to maximal exercise, submaximal and prolonged exercise\textsuperscript{24,28} or similar 16.1km time-trials to that above, but under conditions of simulated altitude of \textasciitilde2500m with consequently reduced exercise intensity,\textsuperscript{29} BC did not facilitate any benefit. Taken together, it appears that benefits of anthocyanin-rich BC supplementation may be restricted to exercise performed at high and very high intensities, in which exercise is restricted to \textasciitilde30mins or shorter durations before task failure occurs.

The above therefore immediately suggests that supplementation with anthocyanin-rich BC extract may facilitate high-intensity exercise performance via the critical power (CP; cycling) or speed (CS; running) concept. This concept relates to exercise with fixed work-rates and assumes that once a threshold (CP or CS) has been exceeded, tolerable duration may be predicted by the specific work-rate being performed. This is because the power- or speed-tolerable duration (S-D\textsubscript{tol}) relationship is fixed to a hyperbolic function that may be accurately calculated, and it therefore also accurately describes the highest sustainable exercise work-rate in the severe intensity domain that may be tolerated for up to a maximum of \textasciitilde30mins before task failure, with the quantity above the threshold measured by achievable work (W'; cycling) or distance (D'; running).\textsuperscript{30-32} Only this work or distance is achievable, because the muscle glycogen and phosphocreatine levels that allow for attaining W’ or D’ fall to limiting values or work is compromised by accumulation of K\textsuperscript{+}, H\textsuperscript{+}, P\textsubscript{i}, adenosine diphosphate, and reactive oxygen species that inexorably disturb intramuscular metabolic and contractile homeostasis.\textsuperscript{30,32} For fixed-speed running, this therefore predicts performance as pacing above the CS will unavoidably cease in a predictable manner and is as such of value for evaluating exercise for exhaustive running up to \textasciitilde30mins.

The aim of this study was therefore to examine the effect of daily anthocyanin-rich BC extract on S-D\textsubscript{tol} during running. The hypothesis was that BC supplementation would right-shift the S-D\textsubscript{tol} in the severe exercise domain and thereby increase CS and D’; i.e. allow for longer-duration exercise at the same speed and/or allow for higher sustainable speed for the same duration of severe-intensity exercise.
Materials and methods

Subjects

15 recreationally active males not engaged in structured training programs volunteered, with characteristics in Table 1. All were health-screened and familiarized to the experiments and signed informed consent forms prior to participation. Exclusion criteria included regular smoking, medication, drugs and pre-existing ergogenic aids, pre-existing medical conditions contraindicative to exercise testing, as well as alcohol and exhaustive exercise within 48 hours. Breakfast was consumed 2hrs before each test, with food and fluids except water avoided after this. The Institutional Review Board approved the study and it conformed to the Declaration of Helsinki.

Experimental design

Subjects attended the laboratory in 2 7-test blocks for intervention and placebo control respectively, with each test approximately at the same time of day (+/-2hrs) and separated by >48hrs and with blocks separated by a 14-day wash-out period, following a double-blind, randomized, placebo-controlled and cross-over design. Subjects warmed-up with a 10-min, 6km/h, 1% gradient treadmill brisk walk/jog (PPS Med. Woodway, Weil am Rhein, Germany). Then, exercise testing commenced with, in order, an incremental exercise test (test 1), assessment of CS and S-Dtol (tests 2-5), and time to exhaustion performance trials (tests 6-7). Subjects were administered 300mg/d BC extract (105mg/d anthocyanin; CurraNZ New Zealand Blackcurrant Extract, Health Currancy, Surrey, UK) or placebo (300mg/d microcrystalline cellulose M102), with n=8/15 starting on BC extract and n=7/15 on placebo, taken daily with breakfast from day -1 and throughout (thus 1-day pre-loading). This regime was adopted due to absorption and plasma anthocyanin peaking 2hrs post-ingestion and clearance within 6-12hrs,3 as well as return to normal status within the wash-out period.33 Nutritionally, 300mg/d BC extract also provided 1kcal/day, while placebo provided 0kcal/d. Subjects were instructed to maintain their normal diet including breakfast and schedule as consistent as possible and received no information of their results until after completing all trials.

Resting measurements

Subjects supine-rested for 10mins before measurements of stature, blood pressure, and blood lactate (La⁻¹) concentration ([La⁻¹]). Body, fat, and lean masses were measured by air-displacement plethysmography (BodPod, Cosmed).
Incremental exercise test

A step-wise ramped incremental exercise test with speed increasing 0.5km/step until volitional exhaustion measured submaximal running economy (CR), La⁻¹ threshold (LT), maximal oxygen uptake (VO₂max), and maximal heart rate (HRmax). Fingerprick capillary blood [La⁻¹] (Analox GM7 Lactate Analyzer, Analox, Hammersmith, UK), oxygen uptake (VO₂) by 1-min Douglas bag-collected expired gas samples (Servomex 4100 Gas Analyser, Servomex, Sussex, UK), and heart rate (HR; Polar Heart Rate S610i, Polar, Kempele, Finland) were measured at end of each step, with step-durations 5mins/step until LT was identified and 1min/step thereafter. LT was determined by the deflection point of La⁻¹ increase, while CR was assessed at 8km/h, LT, and 10km/h. Energy expenditure and substrate utilization rates were estimated at LT by indirect calorimetry, and VO₂max was considered achieved when at least 3 of the following 4 criteria were met: VO₂ plateau, respiratory exchange ratio >1.15, post-exercise [La⁻¹] >8mM, or HR within 10bpm of age-predicted maximum.

Speed-tolerable duration

The S-Dᵦₒˡ relationship was estimated from at least 4 randomized and individualized 1%-inclined treadmill constant speed-tests, each separated by >48hrs and designed to induce volitional exhaustion and exercise intolerance within ~3-20mins (Figure 1A). CS (intercept) and D’ (slope) as parameters for S-Dᵦₒˡ were calculated by least squares linear regression of the linear S-Dᵦₒˡ⁻¹ relationship (Figure 1B):

\[ S = \left( \frac{D'}{t_{tol}} \right) + CS \]

Acceptable limits of the S-Dᵦₒˡ relationship were defined by the standard error (SE) of the estimate: CS <2% and D’ <10%. VO₂ and HR were measured when subjects were considered close to exercise intolerance, while [La⁻¹] was measured 2mins post-cessation, as described above.

Time to exhaustion

As an external test of exercise capacity and performance within the S-Dᵦₒˡ relationship and as such of the validity of the S-Dᵦₒˡ relationship after it had been individually established, time to exhaustion was empirically assessed at the extremes of the S-Dᵦₒˡ relationship, by constant running speeds corresponding to 1) CS, and 2) 110% of that eliciting VO₂max, individualized to each subject as representations of speeds in the extreme lower and upper ends of the S-Dᵦₒˡ relationship. VO₂, HR, and [La⁻¹] were measured close to exercise intolerance, as described above. As a final
test, time to exhaustion trials were repeated after supplementation of 900mg/d BC (315mg/d anthocyanin), administered as described above.

Statistics

The Shapiro-Wilk test confirmed normal distribution. BC versus placebo effects were assessed by paired samples t-tests, and where applicable, one-way analysis of variance (ANOVA), while serial recordings were assessed by repeated measures general linear model; Scheffe post-hoc tests identified effects. Statistical significance was assessed by p<0.05 and the 95% confidence interval of the difference, whereas Cohen’s d reported effect size. Data are expressed as means±standard deviation (SD). Statistical analysis was conducted using SPSS version 26 (IBM, Armonk, NY).
Results

Subject characteristics

Subject characteristics are presented in Table 1, indicating a healthy, young, recreationally active male cohort. Body composition, fitness and blood pressure were within a healthy range and did not change with BC extract supplementation; systolic and diastolic blood pressures after BC supplementation were 121.4±5.5mmHg (p>0.05 [-3.1-2.5] vs placebo) and 82.2±5.9mmHg, respectively (p>0.05 [-1.4-2.7] vs placebo).

Speed-tolerable duration

From the constant speed-tests, the S-D\textsubscript{tol} relationship was plotted for each subject, with tolerable duration well described as a hyperbolic function of speed (example traces in Figure 1A) and CS and D’ calculated after linear S-D\textsubscript{tol}^{-1} transformation (example traces in Figure 1B), with the SE well within the acceptable limits of <2% for CS (range 0.4-1.6%) and <10% for D’ (range 2.0-8.3%).

BC extract supplementation effects on CS and D’ are represented in Figures 1C and D, respectively, with no statistical differences occurring between BC and placebo for either CS (p>0.05 [-0.3-0.1]) or D’ (p>0.05 [-39.1-124.6]). Further analysis however indicated a weak, but statistically insignificant trend toward D’ increasing in 60% of subjects by a magnitude of 10-40% (p=0.08, Cohen’s d=0.17 in those that indicated a trend), suggesting a possibility that certain cohorts may potentially gain a small benefit from BC supplementation. Order of intake of BC extract or placebo did not correlate with the trend toward increased D’ in the aforementioned 60% of subjects (r\textsuperscript{2}=0.106, p>0.05). A similar trend for CS was not observed.

VO\textsubscript{2} and HR measured when subjects were considered close to exercise intolerance during each constant speed-test confirmed volitional exhaustion and attainment of VO\textsubscript{2max} and HR\textsubscript{max} in each test, as expected and required of a valid test.\textsuperscript{30-32} Comparisons between VO\textsubscript{2} measured during the constant speed-tests and the incremental exercise test under both placebo and BC conditions demonstrate that no differences occurred (p=0.981; Figure 1E). Similar effects were reproduced when measuring HR during incremental exercise and constant speed-tests for either of placebo (incremental exercise test 194±6bpm vs constant speed-tests 186±8bpm, p>0.05) or BC (incremental exercise test 191±5bpm vs constant speed-tests 187±6bpm, p>0.05) conditions, with no differences occurring (data not shown); further confirming that volitional exhaustion and exercise intolerance was objectively achieved during each test.
Measurements of post-exercise blood \([\text{La}^-]\) showed a decline from fastest to slowest constant speed-tests (range 11.5-4.4mM), but with no differences occurring between placebo and BC \((p>0.05 [-0.7-0.7]; \text{data not shown})\).

**Time to exhaustion**

As an empirical test of the effect of BC extract supplementation on the S-D_{tol} relationship, we measured time to exhaustion at two fixed speeds, one corresponding to the calculated CS and another corresponding to an exercise intensity that would elicit exercise intolerance within the steeper, upper end of the hyperbolic S-D_{tol} relationship.

At a running speed corresponding to CS (Figure 2A), time to exhaustion ranged 25:40-38:04 min:sec (1540-2284s), whereas at a running speed within the steep upper section of the S-D_{tol} relationship, a speed corresponding to 110% of that eliciting VO_{2max} (Figure 2B), time to exhaustion ranged 3:05-6:07 min:sec (185-367s). However, time to exhaustion did not differ between placebo and BC supplementation at either running speed (CS-speed \(p>0.05 [-82.1-121.8]\) and 110% of VO_{2max}-speed \(p>0.05 [-13.8-28.1])\), though further analysis indicated a weak, but statistically insignificant trend toward time to exhaustion increasing in 60% of subjects by a magnitude of 10-20% \((p=0.09, \text{Cohen’s } d=0.59 \text{ and } 0.42 \text{ in those that indicated a trend for CS-speed and 110% of VO}_{2max}-\text{speed, respectively})\), in line with the previous suggestion that certain cohorts may potentially benefit from BC supplementation. As for D’, order effects BC extract versus placebo did not correlate with the trend toward time to exhaustion \((r^2=-0.065, p>0.05)\). As a final assessment, we repeated the time to exhaustion trials, but after supplementation with 900mg BC (3x normal dose). This did not affect time to exhaustion performance (Figure 2A,B).

In line with the constant speed-tests used to establish the S-D_{tol} relationship, measurements of VO_{2} and HR confirmed that volitional exhaustion and exercise intolerance was objectively achieved at each test by attainment of VO_{2max} and HR_{max} (data not shown). Post-exercise blood \([\text{La}^-]\) also reached the expected elevated levels (range 5.9-15.5mM); however, no differences occurred between placebo and BC \((p>0.05 [-1.4-3.1]; \text{data not shown})\).

**Aerobic capacity, lactate threshold and running economy**

Next, we compared results from the incremental exercise tests. Neither of VO_{2max} (Table 1 and Figure 2C; \(p>0.05 [-0.9-2.1])\) or HR_{max} (Table 1; BC supplementation 191±5bpm; \(p>0.05 [-4-4])\) differed significantly between BC extract supplementation and placebo.
LT, measured as running speed (Figure 2D) at the point of blood La− accumulation, did not significantly differ between BC extract supplementation and placebo ($p>0.05 \ [0.6-0.6]$). LT speed was notably below CS (~9.5 vs ~12km/h; $p<0.05 \ [2.0-2.9]$) under both placebo and BC conditions. LT assessed by VO₂ (Figure 2E) did also not differ significantly between BC supplementation and placebo ($p>0.05 \ [-1.8-1.6]$), and similarly, nor did LT assessed by HR ($p>0.05 \ [-4-3]$; data not shown). VO₂ at LT corresponded to 63 and 64% of VO₂max after placebo and BC supplementation, respectively, whereas HR at LT corresponded to 76% and 74% of HRmax after placebo and BC supplementation. As a result of neither speed or VO₂ at LT differing between placebo and BC supplementation (Figure 2D,E), Cr at LT, expressed as oxygen cost of running (mL·kg⁻¹·km⁻¹), did also not differ between placebo and BC supplementation ($p>0.05 \ [-4.8-6.5]$).

To further study Cr, we recorded steady-state oxygen cost at fixed submaximal running speeds 8 and 10km/h, corresponding to <LT and >LT (LT 9.5±1.6km/h), respectively (Figure 2F). This showed that Cr improved by 3% ($p<0.05 \ [-8.3--5.8]$) when increasing running speed from 8km/h to 10km/h, but did not differ between placebo and BC extract supplementation ($p>0.05 \ [-4.1-4.4]$).

**Energy expenditure and substrate utilization**

Energy expenditure and substrate utilization were assessed during steady-state submaximal exercise conditions at an exercise intensity corresponding to LT (Table 2). This showed that no differences occurred between placebo and BC extract supplementation. Intensities above LT and within the S-Dtol relationship were not assessed, since they are not performed during steady-state conditions.
Discussion

This is the first study to assess the effect of daily supplementation with anthocyanin-rich BC extract on the S-Dtol relationship and its parameters CS and D’ during running. Previous studies had indicated that BC supplementation may improve high-intensity exercise capacity and performance;10,22,25-27 exercise that may at least partly be determined by S-Dtol.30-32 However, in our study, we found no such effect, as our results indicate that the previously reported improvements to high-intensity exercise do not arise via improvements to CS, D’, and the S-Dtol relationship. Although we did note a trend toward increased D’ and time to exhaustion in a cohort of our subjects, these were weak trends that did not reach statistical significance and the effect sizes were small to medium, but variable. Intriguingly though, these were not order effects and are in line with previous findings25 of potential cohort effects, but neither our nor previous studies have been able to identify characteristics within those cohorts sufficient to prospectively target responders vs non-responders for BC supplementation, although it has been noted that benefits of BC supplementation may depend on training history26 and ethnicity.28

Our main conclusion relies on establishment of the S-Dtol relationship. A criticism of the concept may be that it is a computation and not a measurement per se, albeit it is based upon at least 4 separate constant speed-tests.31 We however note that all were deemed valid by close mathematical and physiologic scrutiny of the limits of acceptable error31 and by attainment of VO2max in each separate test due to the slow component of VO2 that inevitably occurs above CS.30-32 Regardless of this, we also empirically measured actual performance within the running speed-intensity range covered by the S-Dtol relationship by subsequent time to exhaustion tests designed to correspond to 1) CS and 2) running speeds in the steep part of the S-Dtol relationship well above CS, where tolerable exercise duration deteriorates rapidly with increasing speeds. Similar to our other results, BC supplementation did not affect time to exhaustion performance, including after subjects received 3x doses of BC supplementation. As such, we conclude that BC supplementation does not alter S-Dtol during running in any predictable manner, and improvements to exercise capacity and performance must therefore originate elsewhere.

High-intensity endurance exercise capacity and performance is also physiologically determined by VO2max, CR and fractional utilization of VO2max during submaximal running, and LT;25 thus, we also included those in our investigation. However, BC supplementation did not affect any of those parameters. This was an expected finding though, based upon previous studies.1,22,28,29
In a departure from comparable previous studies and studies finding upregulation of lipid metabolism genes after cellular exposure to anthocyanins, we found no effect of BC supplementation on substrate utilization and fat oxidation during submaximal exercise corresponding to ~65% of VO\textsubscript{2max}. It would have been interesting to assess fat oxidation during higher-intensity workloads within the S-D\textsubscript{tol} relationship, but this was not feasible in our study since the measurement arrives from steady-state gas exchange recordings, conditions which due to the VO\textsubscript{2} slow component were not present at these intensities. Fat oxidation is however not substantial during high-intensity exercise, whereas in contrast carbohydrate oxidation is, but for the same reason could not be estimated during high-intensity exercise.

We acknowledge that in our study, supplementary BC administration differed from most studies with respect to pre-loading, but not dose, daily time of intake, time prior to testing, or intake with other food. Specifically, most studies have applied a 7-day phase-in period for supplementation, whereas in contrast we applied a 1-day phase-in period. This is because very recent studies have shown that plasma concentration of anthocyanins peaks at 2hrs and clears within 6-12hours post-consumption after intake in the 0.8-3.2mg/kg range, which is a range that also covers our administration of 1.3-1.5mg/kg (105mg/d) anthocyanin, and which therefore suggests that anthocyanin bioavailability is unlikely to differ between the reported short-term 1-7-day pre-loading regimens. However, it remains possible that long-term anthocyanin metabolite build-up may affect the outcome on specific parameters, such as the observation that 5-week daily preloading alleviated markers of post-exercise oxidative stress and inflammation. Nonetheless, administration in our study was at the low end vs other studies with respect to pre-loading and dosage and may have accounted for the lack of significant beneficial effects. However, as noted above, we administered a similar daily dose as most studies that have reported a beneficial effect, and furthermore, increasing dosage to 900mg/d BC (315mg/d anthocyanin) did not cause an effect, which is comparable to previous work reporting no dose-dependent effects on VO\textsubscript{2} or work economy, albeit dose-dependent cardiovascular and substrate oxidation effects have been noted during supine rest and sub-maximal exercise. It also remains possible that mode of intake may affect the outcome, but juice or dry powder-based supplements have not proven more successful. Finally, and as detailed above, we sought to minimize confounding factors, but some day-to-day variation in e.g. subject motivation or nutrition and energy intake consumed during breakfast cannot be excluded; but if they were present, they may have added ecological validity.
As such, we return to the question: does supplementary BC consumption enhance exercise capacity and performance and should it be promoted as an ergogenic aid? Our study cannot find evidence of BC supplementation enhancing high-intensity exercise capacity and performance or enhancing the underlying physiologic parameters that govern high-intensity exercise, but nor do we find evidence of any detrimental effects. Thus, we cannot warrant the use of BC supplementation as an ergogenic aid, but we acknowledge that some,\textsuperscript{8,10,11,17,21-27} but not all\textsuperscript{24,28,29} studies have indicated a positive ergogenic effect. Reasons for this discrepancy remain unknown, but it is possible that administration of BC supplementation must be concurrent to a high training load before a benefit may be realized, given that BC supplementation in some studies has facilitated recovery between exercise sessions,\textsuperscript{25-27} possibly linked to enhanced La\textsuperscript{-} handling with reduced production and/or increased clearance\textsuperscript{24} and tolerance,\textsuperscript{22} reduced oxidative stress and inflammatory immune reactions,\textsuperscript{2-4,8,9,39} and reduced muscle damage\textsuperscript{17} following high-intensity exercise.

**Conclusion**

We could not detect any beneficial or otherwise effect of daily anthocyanin-rich commercially available BC extract supplementation to the S-D\textsubscript{tol} relationship during running, including its parameters CS and D’ or associated time to exhaustion trials as empirical tests of the S-D\textsubscript{tol} relationship, and nor could we detect any effect on VO\textsubscript{2max}, CR and fractional utilization of VO\textsubscript{2max} during submaximal running, or LT. Hence, BC extract supplementation in our study did not demonstrate benefit to high-intensity exercise capacity or performance.
Acknowledgements

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Disclosure statement

The authors report no conflict of interest.
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## Tables

### Table 1: Subject characteristics (n=15).

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
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<tr>
<td>Age (years)</td>
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<tr>
<td>Height (m)</td>
<td>1.76±0.06</td>
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<tr>
<td>Body mass (kg)</td>
<td>74.3±6.7</td>
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<tr>
<td>Body fat (%)</td>
<td>10.8±3.5</td>
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<tr>
<td>Fat mass (kg)</td>
<td>8.3±2.4</td>
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<tr>
<td>Lean mass (kg)</td>
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<td>Body mass index (kg·m⁻²)</td>
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<tr>
<td>VO₂ max (mL·kg⁻¹·min⁻¹)</td>
<td>53.1±3.4</td>
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<tr>
<td>HR max (bpm)</td>
<td>194±6</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>119.1±6.1</td>
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<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>82.7±7.0</td>
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</table>

Data are mean±standard deviation (SD). VO₂ max: maximal oxygen uptake; HR max: maximal heart rate. Values are from placebo, and did not change with blackcurrant extract (BC) supplementation (p>0.05).
Table 2: Energy expenditure and substrate utilization during exercise at lactate threshold (n=15).

<table>
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<tr>
<th></th>
<th>Placebo</th>
<th>BC</th>
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<tr>
<td>Energy expenditure (kJ·min⁻¹)</td>
<td>58.64±5.14</td>
<td>56.2±6.55</td>
</tr>
<tr>
<td>CH oxidation (g·min⁻¹)</td>
<td>2.26±0.61</td>
<td>2.15±0.41</td>
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<tr>
<td>Fat oxidation (g·min⁻¹)</td>
<td>0.59±0.26</td>
<td>0.56±0.44</td>
</tr>
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</table>

Data are mean±standard deviation (SD). BC: supplementation with anthocyanin-rich blackcurrant extract; CH: carbohydrate; No differences occurred between placebo and BC (p>0.05).
Figure captions

**Figure 1**: Characterization of the Speed-tolerable duration (S-D\textsubscript{tol}) relationship during running after placebo or supplementation with anthocyanin-rich blackcurrant extract (BC): A: example traces of the hyperbolic nature of the S-D\textsubscript{tol} relationship; B: example traces of quantification of critical speed (CS; intercept) and D’ (slope) as parameters for S-D\textsubscript{tol}; C: critical speed (CS); D: D’; E: mean and ±1 standard deviation (SD) of maximal oxygen uptake (VO\textsubscript{2max}) measured during incremental exercise test (full and dotted horizontal lines), and oxygen uptake (VO\textsubscript{2}) measured close to exercise intolerance during each constant speed-test (bars). VO\textsubscript{2max} was attained in each constant speed-test. No differences occurred between placebo and BC or between incremental exercise test and constant speed-tests (p>0.05).

**Figure 2**: Time to exhaustion during fixed running speeds after placebo or supplementation with anthocyanin-rich blackcurrant extract (BC) at normal dose (300mg New Zealand Blackcurrant Extract, 105mg anthocyanin) or high dose (900mg New Zealand Blackcurrant Extract, 315mg anthocyanin): A: at critical speed (CS); B: at 110% of maximal oxygen uptake (VO\textsubscript{2max}). Aerobic capacity measured by incremental exercise testing; C: VO\textsubscript{2max}; D: running speed at lactate threshold (LT); E: oxygen uptake (VO\textsubscript{2}) at LT; F: running economy (C\textsubscript{R}) at 8km/h and 10km/h. No differences occurred between placebo and BC (p>0.05). Statistically significant difference 8km/h vs 10km/h: * p<0.05.