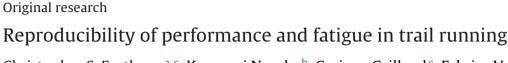
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Christopher S. Easthope^{a,c}, Kazunori Nosaka^b, Corinne Caillaud^c, Fabrice Vercruyssen^d, Julien Louis^a, Jeanick Brisswalter^{a,*}

^a Laboratory of Human Motricity, Sport, Education and Health, University of Nice Sophia Antipolis, France

^b School of Exercise and Health Sciences, Edith Cowan University, Australia

^c School of Exercise and Sport Science, University of Sydney, Faculty of Health Science, Australia

^d Laboratory of Human Motricity, Sport, Education and Health, University of South France Toulon-Var, France

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ABSTRACT

Objectives: This study aimed to test the reproducibility of running performance, neuromuscular fatigue markers and indirect muscle damage indicators in a field-based trail time-trial.

Design: Running performance and changes in classical physiological parameters were analysed in 11 experienced trail runners before and in the days following four bouts of outdoor trail running (15.6 km), 7 days apart.

Methods: Heart rate, running time and lactate concentration were monitored in each running bout. Maximal voluntary contraction torque, counter movement jump height, plasma creatine kinase activity and muscle soreness were assessed before and 1, 24 and 48 h post-race. Within-bout changes were elucidated using a two-way repeated measures ANOVA. Inter-repetition reproducibility was examined using an intraclass correlation coefficient (R) and the mean intra-subject coefficient of variation at each measurement time point.

Results: Running time was longer (p < 0.05) for the first bout compared with the other three bouts. Magnitude and time course of changes in counter movement jump height, creatine kinase activity and muscle soreness were similar among all four bouts (overall peak means: -17%, +35% and 54/100 mm respectively). The acute reduction in maximal voluntary contraction torque (peak mean: -17%) was attenuated exclusively in the fourth bout (p < 0.05). The two middle bouts showed good reproducibility (intraclass correlation coefficient and coefficient of variation) for running time, maximal voluntary contraction torque and counter movement jump height, but low to moderate for creatine kinase activity, muscle soreness, blood lactate and rate of perceived exertion.

Conclusions: A short outdoor trail run is a reliable model for investigations of fatigue and muscle damage, but certain methodological precautions should be respected.

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1. Introduction

Trail races are off-road endurance runs covering distances from 15 to 75 km (>90 km for ultra trails) on unsurfaced mountain trails with extensive vertical displacement.¹ Distance and the climb distance⁻¹ ratio (E/D, normal range: 40–65 m km⁻¹; 8–13%) are the main performance parameters.^{1,2} Recent studies investigating trail races reported aspects of neuromuscular fatigue mainly assessed by maximal voluntary contraction (MVC) torque and changes in twitch and activation parameters.^{1–3} For example, MVC torque of the knee extensors has been reported to decrease 23.5% after a 30 km trail run,⁴ 32% after a 55 km trail run¹ and 35% after a 166 km mountain ultra-marathon.² The neuromuscular fatigue is often accompanied by increases in self-reported muscle soreness ratings and plasma bulk damage markers, such as creatine kinase (CK),^{1,2,5} lasting for several days. This is associated with an exacerbated eccentric component invoked in the downhill phases. The physiological stress profile elicited through combined fatigue and muscle damage is specific to trail running.

Participant increases⁶ invite the investigation of and development of strategies to minimise neuromuscular fatigue and structural damage to the muscle. However, evaluating trail-specific interventions is challenging, as trail race simulation in a laboratory is difficult due to terrain and grade variability. It may therefore be more effective to assess strategies and modalities that could affect performance and recovery in a field setting. Under this constraint, two factors might affect reproducibility of parameters examined in field trail runs.

* Corresponding author. E-mail address: brisswalter@unice.fr (J. Brisswalter). Firstly, studies conducted in the field are associated with higher variability induced, for example, through environmental factors







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(temperature, wind, humidity and surface conditions). While the test-retest reproducibility of treadmill-based protocols has been frequently evaluated,^{7,8} no previous study has investigated the reproducibility of variables associated with running performance, neuromuscular fatigue and muscle damage in a field-based trail run.

Secondly, it is well known that an initial bout of eccentric exercise induces a protective effect which decreases muscle damage and ameliorates recovery in subsequent bouts. This protective effect is referred to as "the repeated bout effect" and is generally observed from 2 to 6 weeks following the initial intervention in untrained muscles.^{9,10} There have been several reports of diminished effect magnitude in trained muscle^{11,12} yet, to the best of our knowledge, no previous study has investigated the repeated bout effect in a trail running model, especially performed by trained runners.

Classical fatigue-induction models are not suited to examining trail running as they do not take into account the rather severe gradients and variable surface encountered on typical courses. Prior trail investigations employed either treadmill simulation, ^{13,14} which disregards the terrain component completely, or competition analysis, ^{1,2,4} which is unsuited to intervention type investigations and involves a complicated measurement set-up. Therefore this study employed a short (<20 km) short distance trail with a medium E/D^{1,2} of 52.88. This model is straightforward to implement and has the additional advantage that it reflects a typical training distance for recreational trail runners and entails a short recuperation time.

The aim of this study was therefore to examine the feasibility of using an outdoor trail run to evaluate future intervention strategies. To this end, the reproducibility of neuromuscular fatigue and structural muscle damage markers over 4 bouts of a 15.6 km trail run was determined in experienced trail runners.

2. Methods

Eleven actively competitive male trail runners (age: 34.7 ± 9.8 years, body mass: 72.3 ± 6.8 kg, height: 178.4 ± 7.0 cm, maximal oxygen uptake: 60.1 ± 6.5 mL min⁻¹ kg⁻¹) participated in this study. Inclusion criteria included a minimum of 2 years trail racing experience and a training volume of 40-100 km wk⁻¹ (mean: 60 ± 20 km wk⁻¹) in the 3 months preceding initial testing. For 2 days before and after each trial, the runners were requested to refrain from exercise and to adhere to a standardised nutritional routine. Written informed consent was obtained and the study was approved by the Institutional Human Research Ethics Committee.

After an initial maximal oxygen uptake (VO2max) test on a treadmill, all participants performed four bouts of trail running on the same course with 7 days rest between bouts. In each bout, running time, heart rate, post-run ratings of perceived exertion and blood lactate concentration were recorded. Immediately before (pre) and 1 (post), 24 and 48 h after the run the following parameters were assessed: maximal voluntary isometric knee extension (MVC) torque, counter movement jump (CMJ) height, plasma creatine kinase (CK) activity and muscle soreness. These variables were examined over time in each bout and each time point was compared between bouts.

Two weeks before the first bout, all participants completed a maximal incremental running protocol on a treadmill (+4%, Gymrol S2500, HEF Tecmachine, Andrezieux-Boutheon, France) in the lab while heart rate (RS800, Polar, Kemple, Finland) and pulmonary gas exchange (Oxycon Alpha, Jaeger, The Netherlands) were recorded. All instruments were calibrated before each test as described by the manufacturers. The protocol consisted of a 6 min warm-up at 9 km h^{-1} followed by an increase of 1 km h^{-1} every 2 min until

volitional exhaustion. Maximal heart rate (HRmax) and oxygen uptake (VO2max) were determined as the highest 30 s mean, fulfilling the classical criteria of a respiratory equivalent greater than 1.1, an HR greater than 90% of the age prediction and a plateau in VO2 despite an increase in mechanical intensity.¹⁵

The trail time-trial consisted of 3 laps of a 5.2 km course (total distance: 15.6 km) starting close to sea level. Each lap was composed of a climbing segment (2200 m, 13%, 275 m climb) followed by a downhill segment (3000 m, -9%, 275 m descent). The course was exclusively on mountain single tracks with repeated technical sections on rocky and root-covered paths. Each participant was weighed and equipped with a Polar RS800 heart rate monitor, 680 ml of fluid containing carbohydrates (74 g L^{-1}) and 2 energy gels (carbohydrates: 18 g gel⁻¹). All participants were asked to wear similar clothes for each bout and to aim for the best completion time possible. Starting times were staggered, allowing 20 min between participants. Immediately after the run, a blood sample was taken from the ear lobe for lactate analysis (Lactate Pro, Arkray, Amstelveen, The Netherlands), participants were weighed, and RPE was verbally queried while standing using standard terminology and a 6–20 point Borg Scale.¹⁶

MVC testing took place in the laboratory about 10 min drive from the time-trial course before and 1, 24 and 48 h after the run. Following the motorised transfer, participants were securely strapped into an isokinetic dynamometer (Biodex System 3, Shirley, New York, USA) with the knee joint angle of the right leg at 90° (full leg extension = 0°). The axis of the knee joint was carefully aligned with the rotational axis of the dynamometer and all settings were kept constant throughout the experiment. Before each MVC, participants warmed up on the isokinetic dynamometer by repeating 10 one-second isometric contractions at 50% MVC (one second rest between contractions). After 3 min rest, in which participants were asked to indicate perceived muscle pain of the knee extensors on a 10 cm visual analogue scale visibly anchoring zero for 'no pain' and 10 for 'maximal pain', testing commenced. Participants were instructed to "extend the knee as hard and fast as possible" for the three 5-second MVC measures (55 s rest between attempts) while standardised verbal encouragement was given. The highest MVC value achieved in the three attempts was used.

Ten minutes after MVC testing, participants were positioned on an Ergo Jump system (Boscosystem, S. Rufina, Italy) and instructed to place their hands on their hips and to jump as high as possible and land with extended legs. Jumping position was standardised as described previously,¹⁷ and the participants practised extensively under supervision before the measurements. Three jumps with 30 s rest between attempts were then recorded. The maximum jump height achieved was used for further analysis.

Blood samples were drawn from the antecubital vein using a standard vacutainer system and centrifuged for 10 min to obtain plasma. Plasma samples were aliquoted and stored in a freezer $(-80 \,^{\circ}\text{C})$ until analysed for CK activity by a Roche Hitachi 911 chemistry analyser (Roche Diagnostics Corporation, Indianapolis, IN, USA).

A two-way repeated measures ANOVA (time (4) × bout (4)) was conducted on the absolute values of MVC torque, CMJ height, plasma CK activity, muscle soreness and lap times. A Newman–Keuls post hoc test was used for multiple comparisons to identify differences between individual time points. Reproducibility of parameters across bouts was examined with an intraclass correlation coefficient (ICC, *R*) and the mean intra-individual coefficient of variation (CV) was calculated for each time point.¹⁸ Reproducibility was judged by the *R* values of ICC¹⁹: 0–0.25: little, 0.26–0.49: low, 0.50–0.69: moderate, 0.70–0.89: high, and 0.9–1.0: very high. The significance level was set at p < 0.05 and all data are presented as means ± standard deviation (SD).

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