European Journal of Applied Physiology (2019) 119:1479–1489
https://doi.org/10.1007/s00421-019-04135-4

ORIGINAL ARTICLE

Stretch–shortening cycle exercise produces acute and prolonged impairments on endurance performance: is the peripheral fatigue a single answer?

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Received: 8 November 2018 / Accepted: 1 April 2019 / Published online: 5 April 2019

Abstract

Objective This study aimed to verify the acute and prolonged effects of stretch–shortening cycle exercise (SSC) on performance and neuromuscular function following a 4-km cycling time trial (4-km TT).

Methods On separate days, individuals performed a 4-km TT without any previous exercise (CON), immediately (ACUTE) and 48 h after (PROL) SSC protocol (i.e., 100-drop jumps). Neuromuscular function was measured at baseline SSC (baseline), before (pre-TT) and after (post-TT) 4-km TT. Muscle soreness and inflammatory responses also were assessed.

Results The endurance performance was impaired in both ACUTE ($-2.3 \pm 1.8\%$) and PROL ($-1.8 \pm 2.4\%$) compared with CON. The SSC protocol caused also an acute reduction in neuromuscular function, with a greater decrease in potentiated quadriceps twitch-force ($Q_{tw.pot}$ $-49 \pm 16\%$) and voluntary activation (VA $-6.5 \pm 7\%$) compared for CON and PROL at pre-TT. The neuromuscular function was fully recovered 48 h after SSC protocol. Muscle soreness and IL-10 were elevated only 48 h after SSC protocol. At post-TT, $Q_{tw.pot}$ remained lower in ACUTE ($-52 \pm 14\%$) compared to CON ($-29 \pm 7\%$) and PROL ($-31 \pm 16\%$).

Conclusion These findings demonstrate that impairment in endurance performance induced by prior SSC protocol was mediated by two distinct mechanisms, where the acute impairment was related to an exacerbated degree of peripheral and central fatigue, and the prolonged impairment was due to elevated perceived muscle soreness.

Keywords Inflammatory response · Central fatigue · Peripheral fatigue · Muscle soreness · Self-paced exercise

Abbreviations

SSC	Stretch–shortening cycle exercise
4-km TT	4-km cycling time trial
PPO	Peak power output
EMG	Electromyographic activity
RMS	Root mean square
RPE	Rating of perceived exertion
VA	Voluntary activation
MVC	Maximal voluntary contraction force
$Q_{tw.pot}$	Quadriceps potentiated twitch-force
DOMS	Delayed onset muscle soreness
CK	Creatine kinase
LDH	Lactate dehydrogenase
IL-1b	Interleukin-1 beta
IL-6	Interleukin-6
IL-10	Interleukin-10
TNF-α	Tumor necrosis factor-α

Communicated by Nicolas Place.

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Introduction

Plyometric exercise has been classified as type of stretch–shortening cycle (SSC) exercise due to active muscle stretch (i.e., eccentric contraction) immediately followed by active muscle shortening (i.e., concentric contraction) (Nicol et al. 2006). It is interesting to observe that plyometric exercises have been increasingly implemented in the training routine of endurance athletes, probably due to the increased number of evidence demonstrating their benefits to the endurance performance (Paavolainen et al. 1999; Paton and Hopkins 2005; Blagrove et al. 2018). However, a consequence of intense eccentric contraction or even low-intensity eccentric contraction during the prolonged time is the appearance of symptoms associated with exercise-induced muscle damage (i.e., swelling, soreness, increased blood myofibre proteins and impairment in muscular function). As a result, a possible negative influence of these exercises on endurance performance may persist for several days, mainly in individuals unaccustomed to this type of exercise (Komi 2000).

Interestingly, the recovery of the capacity of the skeletal muscle to produce maximum force after SSC exercise presents a bimodal behavior, where occurs an immediate reduction, quick recovery within 1–2 h, followed by a secondary impairment on the following 1–2 days (Nicol et al. 2006). Both the initial and the delayed reductions in capacity of skeletal muscle to produce maximum force, often measured by maximum voluntary isometric contraction (MVC), have been attributed to myofibrillar disruption (Hortobagyi et al. 1998). It is important to note that the secondary reduction in muscular function seems to be due to exacerbates the initial muscle damage caused by increased muscle degradation triggered by an increase in the intracellular calcium concentration and/or inflammatory response (Clarkson and Sayers 1999). It suggests that the acute and prolonged reduction in the capacity of skeletal muscle to produce maximum force (i.e. neuromuscular fatigue) after intense SSC seems to be caused by intramuscular alterations (i.e. peripheral fatigue). However, central modulation seems to occur with the achievement of intense SSC. In fact, studies, using the twitch interpolation technique, have found a reduction in voluntary activation (a marker of central fatigue) immediately and days after SSC. It is been suggested that a reduction in voluntary activation may be caused by an inhibitory effect induced by muscle soreness, swelling, and stiffness (Byrne et al. 2004). Furthermore, the increased concentrations of cytokines would modulate neural activity in the central nervous system through stimulation of vagus nerve and/or the blood–brain barrier, which in turn can act to influence the circumventricular organs in the brain (Vargas and Marino 2014).

Based on previous findings showing that a plyometric exercise protocol composed of 100 drop jumps induces neuromuscular fatigue without producing significant increases in muscle metabolites (i.e., Pi, PCr, ATP, lactate) (Nielsen et al. 2005), some studies have used this methodological approach as a tool to test a relatively new fatigue model (i.e. psychobiological model) that do not consider intramuscular metabolic perturbation as an important factor for endurance performance (de Morree and Marcora 2013; Marcora et al. 2008). In general, these studies have reported that acute and delayed reductions in high-intensity endurance exercise performance promoted by previous intense SSC were accompanied by a greater perception of effort (Marcora et al. 2008; de Morree and Marcora 2013) and a reduced neural drive to the exercised muscle during the endurance task (Marcora and Bosio 2007; Burt and Twist 2011), respectively. This has led some authors to suggest that endurance performance is limited by a more task disengagement during constant-load exercise or increased perception of effort during time trials rather than exacerbation of the peripheral and/or central fatigue (de Morree and Marcora 2013; Marcora 2010; Marcora et al. 2008). However, this explanation remains a conjecture since these studies have been limited to perceptual responses (perceived effort and DOMS) or indirect markers of the central motor command (as inferred by electromyography and/or heart rate) during the endurance task (de Morree and Marcora 2013; Marcora et al. 2008; Marcora and Bosio 2007; Burt and Twist 2011).

Therefore, the aim of the present study was to evaluate the contribution of central and peripheral processes for the neuromuscular fatigue induced by a 4-km TT performed immediately and 48 h after the SSC protocol. We hypothesized that (1) acute impairment in 4-km TT performance would be mainly due to exacerbated, pre-exercise peripheral fatigue; (2) central fatigue would be increased when 4-km TT is performed 48 h after SSC protocol.

Methods

Participants

The sample size required was estimated using G*Power software (version 3.1.9.2), with data from a previous investigation that analyzed the effect of intense SSC (i.e., 100-drop jumps) on neuromuscular function (Skurvydas et al. 2002). A sample size of six participants was estimated to achieve statistically significant differences in MVC, for an expected effect size of 1.56 and power of 0.8 with an alpha level of 0.05. Therefore, eight male cyclists (age 34 ± 7 years, body mass 76 ± 12 kg, height 179 ± 10 cm, body fat 14 ± 5%, peak power output (PPO) 389 ± 52 W, and maximal oxygen uptake 54 ± 6 ml kg^{-1} min^{-1}) participated in this study.

Participants were classified as recreationally trained cyclists in accordance with De Pauw et al. (2013). This investigation was approved by the local ethics committee and was conducted in accordance with the Declaration of Helsinki.

Experimental design

Each participant visited the laboratory on six occasions (Fig. 1). On the first visit, they performed a maximal incremental test (150 W + 30 W every 1 min) on a cyclosimulator (RacerMate®, Computrainer™, Seattle, Washington, USA) to determine their $\dot{V}O_{2max}$ and PPO to characterize the participants. Thirty minutes after the incremental test, participants performed the first 4-km TT familiarization. In the second and third visits, participants performed two other familiarizations with the 4-km TT and all experimental procedures. During the following three visits, participants performed a 4-km TT under the following experimental conditions: (1) immediately after 100-drop jumps (ACUTE); (2) 48 h after ACUTE (PROL); (3) without previous drop jump protocol (CON). The CON and ACUTE were performed in a counterbalanced crossover order. A minimum interval of 7 days after PROL was chosen to avoid the effects of residual fatigue on the control condition. All experimental trials were performed in the morning to avoid the impact of circadian variation (Fernandes et al. 2014). The participants were asked to refrain from vigorous physical activities, caffeine, and alcohol 24 h before each experimental session.

Experimental trials

All trials were performed using the participant's own bike attached to a cyclosimulator (RacerMate®, Computrainer™, Seattle, WA, USA). Before each trial, the cyclosimulator was calibrated in accordance with the manufacturer's recommendations. Before the 4-km TT, the participants underwent a warm-up at 150 W with 90 rpm for 5 min. Then, participants were instructed to perform the 4-km TT in the shortest possible time. They were free to change gear and pedal frequency as desired during the entire trial. Feedback only from the distance covered was provided continually. The power output was recorded at a rate of 1 Hz, stored in Computrainer software (RacerMate Software, Version 4.0.2, Seattle, WA, USA) and, subsequently, averaged over 1000 m. The electromyographic activity (EMG) were measured during the entire 4-km TT. The rate of perceived exertion (RPE) was obtained every 500 m using the Borg 15-point scale.

Stretch–shortening cycle protocol

The SSC protocol consisted of ten sets of ten consecutive maximal drop jumps, interspersed with a 1-min recovery between each set. Previous studies have demonstrated that a similar protocol was able to effectively induce acute (de Morree and Marcora 2013; Marcora et al. 2008) and prolonged (48-h post) neuromuscular fatigue (Marcora and Bosio 2007). Prior to starting the SSC protocol, a maximal drop jump was performed and the height was measured using a contact mat (Jump System Pro, Cefise, São Paulo, Brazil). In short, the participants drooped 100 times from a 40-cm platform down to a 90° knee angle before jumping upward keeping the target height for each jump. During the entire test, the jump performance was provided to the participant in real-time visual feedback via a computer screen. All jumps were performed with hands on hips.

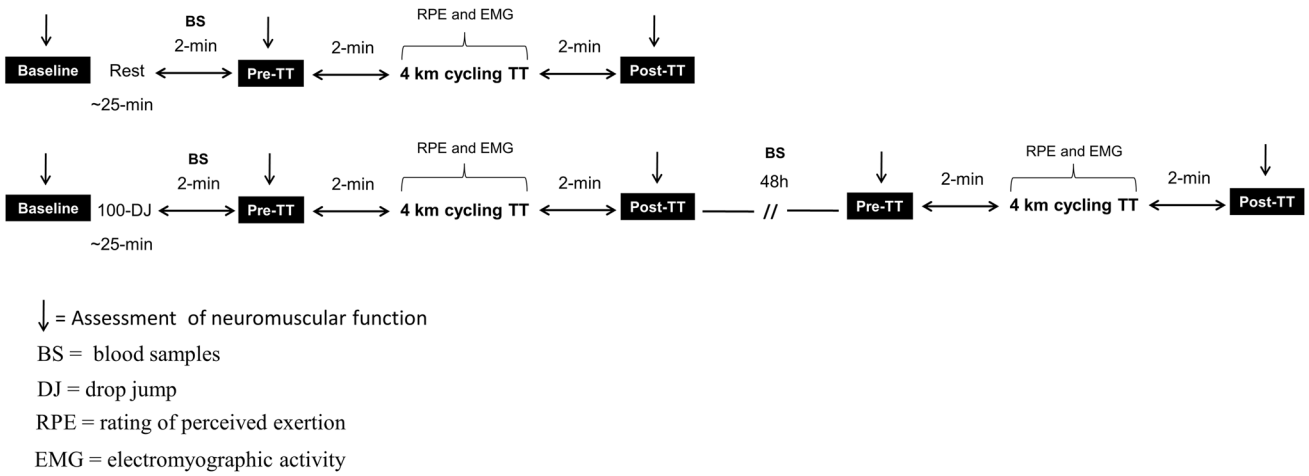

Fig. 1 Schematic illustration of the experimental protocol

Blood sampling, biochemical markers of muscle damage, and inflammatory response

Blood samples (~ 10 ml) were collected from a superficial forearm vein with participants seated immediately before the 4-km cycling TT using EDTA Vacutainer Tubes. Then they were centrifuged at 4000 revs. min^{-1} for 20 min, at 4 °C, to separate the plasma. This plasma was stored in multiple aliquots at − 80 °C for posterior analysis. The CK and LDH activities were determined in plasma using a UV spectrophotometer (EONC, Biotek Instruments, Winooski, VT, USA) and specific kit (Bioclin, Belo Horizonte, MG, Brazil). Plasma concentrations of interleukin-1 beta (IL-1b), interleukin-6 (IL-6), interleukin-10 (IL-10), and tumor necrosis factor-α (TNF-α) were analyzed in duplicate using Bio-Plex Pro (Bio-Rad Laboratories, Hercules, CA). The intra-assay coefficients of variation for IL-1b, IL-6, IL-10, and TNF-α were 3.6, 6.8, 5.8, and 4.3%, respectively.

Perceived muscle soreness

The DOMS was subjectively assessed under each experimental condition using a seven-point Likert scale (Vickers 2001). The participants were asked to rate the overall level of delayed-onset muscle soreness felt in both legs during the previous 12 walking hours according to the following verbal anchors: (0) a complete absence of soreness; (1) a light pain felt only when touched/a vague ache; (2) a moderate pain felt only when touched/a slight persistent pain; (3) a light pain when walking up or down stairs; (4) a light pain when walking on a flat surface/painful; (5) a moderate pain, stiffness or weakness when walking/very painful; and (6) a severe pain that limited the ability to move.

Neuromuscular function

Force measurements

In order to measure voluntary and evoked forces by the quadriceps muscles, participants sat on a chair (Cefise, Nova Odessa, SP, Brazil) equipped with a load cell (linear range 0–2224 N; sensitivity 2 mV V^{-1} and 2.27 mV N^{-1}) (SML-500, Interface, Scottsdale, AZ, USA). The force produced by the knee extensor muscles of the right leg was sampled at 1000 Hz using an A/D converter interfaced with a computer using MyoResearch software version 1.08.38 (Noraxon, Scottsdale, AZ, USA). The participants kept the trunk angle set at 120° and knee joint angle set at 90°. A noncompliant strap connected to a load cell was attached around the subject's ankle. The participants were firmly secured to the chair with noncompliant straps to minimize body movement. Before the MVC, the participants performed five 5-s contractions of the knee extensors, interspersed by 30-s rest periods, at intensities corresponding to 50, 60, 70, 80, and 100% of the self-perceived maximal force. Thereafter, participants performed three 5-s MVC with 30-s rest between them. Recommendations previously suggested (Gandevia 2001) related to instruction, practice, visual feedback of performance, and standardized verbal encouragement were adopted in the present study to ensure the maximum effort of participants during the MVCs. Furthermore, participants were familiarized with the force measurements and evoked twitch responses on three different days (visit 1, 2 and 3).

Electrical motor nerve stimulation

Evoked forces of the knee extensor muscles were induced by the electrical stimulus of the femoral nerve (Neuro-TES, Neurosoft, Ivanovo, OI, Russia). A monopolar 0.5-cm diameter cathode self-adhesive electrode (Ambu Neuroline 715, Ballerup, Denmark) was placed on the femoral nerve and a monopolar 0.5-cm diameter anode self-adhesive electrode (Ambu Neuroline 715, Ballerup, Denmark) on the gluteal fold opposite to the cathode. The thigh of each subject was mapped for the position of the electrodes using anatomical reference points (i.e., the border of patella) and skin marks (i.e., freckles, and scars) via transparent plastic. The optimal intensity of the electrical stimulation was determined by increasing the voltage until the twitch torque and M-wave amplitude had reached a plateau. The plateau was identified when no further increase in twitch torque and M-wave amplitudes was observed despite three consecutive increases in intensity. The stimulation intensity used in all tests was supramaximal (120% of the plateau) (Neyroud et al. 2014). Similar to previous studies (Amann and Dempsey 2008; Amann et al. 2009) single twitch was superimposed when the plateau of the MVC was reached and 2 s after the MVC (relaxed muscle) to obtain quadriceps potentiated twitch-force ($Q_{tw.pot}$). The electrical motor nerve stimulation set (i.e. superimposed and potentiated twitch) was repeated three times, with a 30-s interval between sets, for each time point. The neuromuscular function was assessed at baseline, before (pre-TT), and 2 min after 4-km TT (post-TT) (Fig. 1).

Maximal voluntary activation

The ratio of the amplitude of the superimposed single twitch over the size of the potentiated single twitch was calculated to obtain voluntary activation (VA) as follows:

$$VA = (1 - \text{superimposed twitch}/\text{potentiated twitch}) \times 100. \quad (1)$$

In some cases, when the superimposed stimuli were applied at the time that the torque level was already slightly declining, a correction was applied to the original equation, as suggested by Strojnik and Komi (1998).

Electromyographic recordings

EMG of the right vastus lateralis muscle was recorded via bipolar Ag–AgCl surface electrode (Hall, São Paulo, SP, Brazil) at an interelectrode distance of 20 mm. The reference electrode was placed on the tibial anterior tuberosity. The skin preparation, placement, and location of the electrodes were in accordance with the recommendations of SENIAM (Hermens et al. 2000). EMG data were collected at a sampling frequency of 2000 Hz (Neuro-MEP-Micro, Neurosoft, Ivanovo, Russia). The raw EMG signal was full-wave rectified and filtered with second-order Butterworth band-pass filters with cutoff frequencies set at 20 and 500 Hz to remove external interference noise and movement artifacts. The root mean square (RMS) of the EMG signal recorded during the entire time trial was normalized to the RMS recorded during MVC at baseline. RMS during each MVC was calculated as the average value over 250 ms before the electrical stimulation. M-wave peak-to-peak amplitude was calculated for each potentiated twitch.

Statistical analysis

Data are reported as mean ± SD. Data distribution was verified by the Shapiro–Wilk's test. The change in neuromuscular function, power output, and EMG during 4-km cycling TT was analyzed using a two-way (trial x time) repeated measures ANOVA. A one-way repeated-measures ANOVA was used for other variables. When the main effect was detected, the multiple-comparisons analysis was performed using Tukey's post hoc test. A statistical level of significance ≤ 0.05 was used for all analysis. Statistical analyses were conducted using Statistica 10.0 (StatSoft, Inc., Tulsa, OK, USA).

Results

Reproducibility measurements

The typical error expressed as coefficient of variation (CV%) was calculated for assessment of the performance (recorded during the second and third familiarization with 4-km cycling TT) and neuromuscular function reproducibility (recorded during the baseline of CON and $ACUTE_{4\text{-km TT}}$ conditions). Coefficient of variation was 1.1 ± 0.7% (0.0–2.1%) for time to complete 4-km cycling TT, 4.2 ± 3.7% (1.9–12.3%) for MVC, 7.1 ± 3.5% (2.6–11.7%) for $Q_{tw.pot}$, and 2.3 ± 1.8% (0.4–5.7%) for VA (Fig. 2).

Neuromuscular function before 4-km TT

The participants showed similar baselines values for MVC (629 ± 81 vs 626 ± 103 N) and perceived muscle soreness (0.8 ± 1.0 vs 0.1 ± 0.4 A.U.) between ACUTE and CON, demonstrating that washout period provides sufficient time for recovery. The percentage of changes from baseline to pre-TT of the global (i.e., MVC), peripheral (i.e., $Q_{tw.pot}$), and central (i.e., AV) fatigue parameters are shown in Fig. 3. The 4-km TT in ACUTE started with a larger reduction

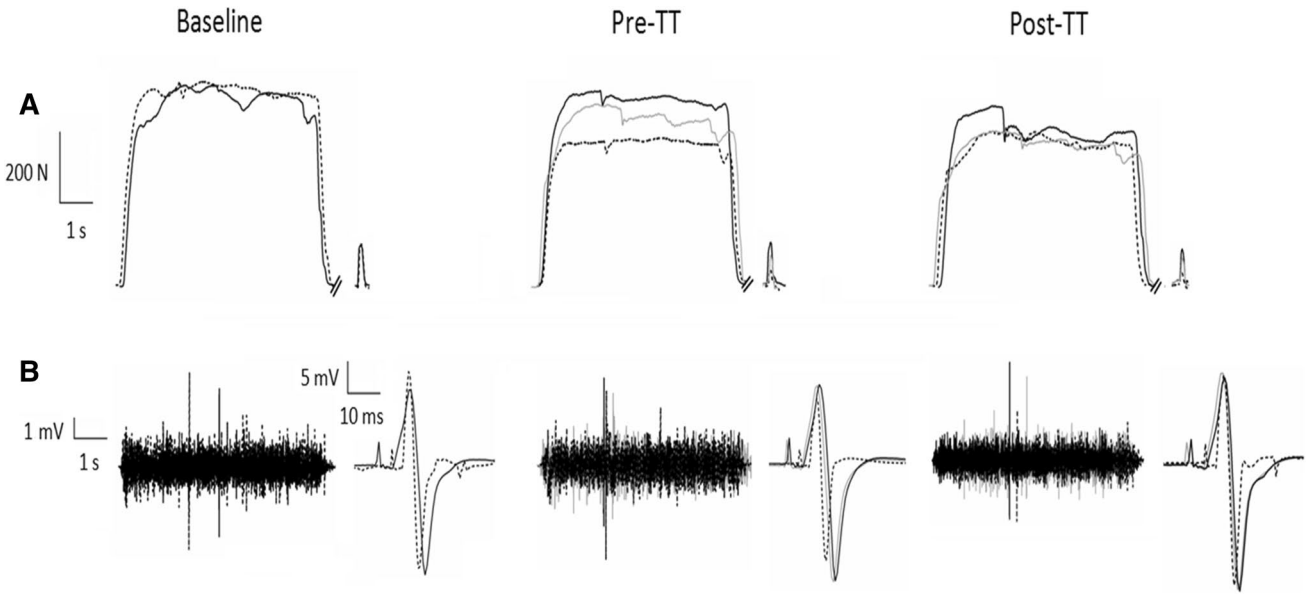

Fig. 2 A typical raw traces of maximal voluntary contraction, superimposed twitch and potentiated twitch (**a**), vastus lateralis electromyographic and M-wave amplitude (**b**) of one participant at baseline, before (pre-TT) and after (post-TT) 4-km cycling time trial in the CON (black line), ACUTE (dashed line) and PROL (grey line)

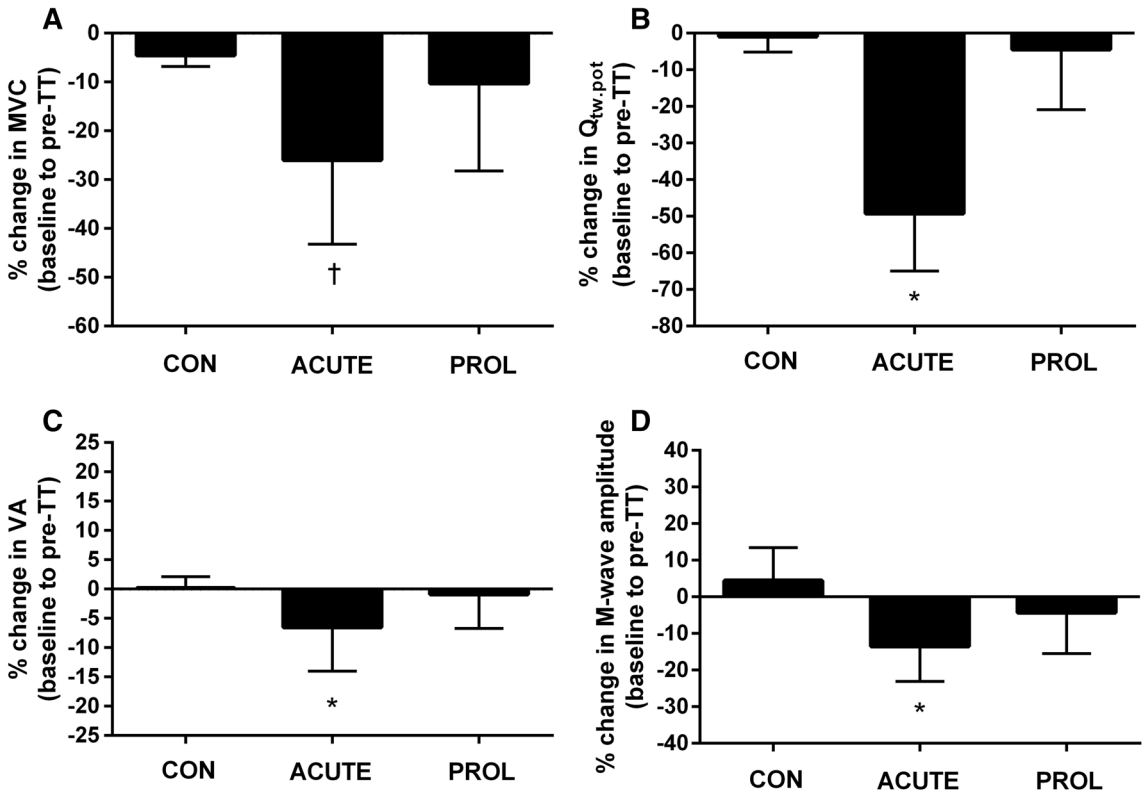

Fig. 3 Percentage of change in neuromuscular function from baseline to before high-intensity endurance exercise. *MVC* maximal voluntary contraction, $Q_{tw.pot}$ potentiated quadriceps twitch-force, *VA* voluntary activation, *M-wave* M-waves amplitude. In the PROL condition, alterations in neuromuscular function were expressed as a percentage of changes from baseline of the ACUTE condition. Data are expressed as mean ± SD. *Significantly different from CON and PROL ($P < 0.05$). †Significantly different from CON ($P < 0.05$)

in MVC ($P < 0.01$), $Q_{tw.pot}$ ($P < 0.01$), VA ($P = 0.04$), and M-wave ($P < 0.01$) (Fig. 3). In contrast, all these variables recovered fully within 48 h in the PROL (Table 1, $P > 0.05$). As expected, there was no significant variation in any variable from baseline to pre-TT for control condition (Table 1).

DOMS, markers of muscle damage and inflammatory responses

The markers of muscle damage and inflammatory responses before experimental conditions are shown in Table 2. The 4-km TT in ACUTE started at similar levels (all $P > 0.05$) of DOMS, CK, LDH, IL-1, IL-6, and TNFα to CON (Table 2). However, in PROL, it started with increased levels of DOMS (Table 2), compared to both CON and ACUTE ($P < 0.01$), and increased levels of IL-10, compared to ACUTE ($P = 0.04$).

Performance, muscle recruitment, and physiological and perceptual responses during 4-km TT

The overall 4-km TT performance was impaired in both ACUTE (− 2.3 ± 1.8%, $P = 0.02$) and PROL (− 1.8 ± 2.4%, $P = 0.05$) compared with CON, but there was no difference ($P = 0.75$) between PROL and ACUTE conditions (Fig. 4). Six out of eight subjects showed reduced endurance performance in the $ACUTE_{4\text{-km TT}}$, while seven out of eight subjects showed reduced endurance performance in the $PROL_{4\text{-km TT}}$ when compared with CON.

Main effects for condition ($P = 0.005$) and distance ($P = 0.001$) were found for power output ($P < 0.05$), but there was no significant interaction ($P = 0.268$). When compared to CON condition, the mean power output was lower in ACUTE and PROL. However, there was no significant ($P = 0.680$) difference between ACUTE and PROL conditions. Central motor command during endurance exercise, as inferred by EMG, was similar across conditions, since only distance effect was found ($P < 0.001$, Fig. 4). Likewise, RPE was similar ($P > 0.05$) between conditions (CON 15.8 ± 1.8, ACUTE 16 ± 1.9 and PROL 15.7 ± 2.3).

Neuromuscular function following 4-km TT

The percentage of changes from baseline to post-TT of the global (i.e., MVC), peripheral (i.e., $Q_{tw.pot}$), and central (i.e., VA) fatigue parameters is shown in Fig. 5. The MVC

Table 1 Neuromuscular responses to electrical stimulation of the motor nerve

	CON	ACUTE	PROL
Global fatigue			
Baseline	629 ± 81	626 ± 103	–
MVC (N)			
Pre-TT	601 ± 76	467 ± 131*	567 ± 164
Post-TT	492 ± 99*,†	460 ± 132*	492 ± 180‡
Central fatigue			
Baseline	94 ± 3	94 ± 2	–
VA (%)			
Pre-TT	94 ± 3	88 ± 7*	93 ± 6
Post-TT	90 ± 7	88 ± 5*	91 ± 8
Peripheral fatigue			
Baseline	168 ± 29	169 ± 27	–
$Q_{tw.pot}$ (N)			
Pre-TT	167 ± 32	87 ± 34*	164 ± 45
Post-TT	120 ± 27*,†	82 ± 31*	117 ± 36‡,†
M-wave amplitude (mV)			
Baseline	23 ± 5	24 ± 4	–
Pre-TT	24 ± 5	21 ± 4*	23 ± 2
Post-TT	27 ± 4*,†	24 ± 4†	26 ± 2†

MVC maximal isometric voluntary contraction force, *VA* voluntary activation, $Q_{tw.pot}$ quadriceps potentiated twitch-force

*Significantly different from baseline ($P < 0.05$). †Significantly different from Pre-TT ($P < 0.05$). ‡Significantly different from baseline in ACUTE ($P < 0.05$)

Table 2 Markers of muscle damage and inflammatory responses

	CON	ACUTE	PROL
DOMS (A.U.)	0.1 ± 0.4	0.8 ± 1.0	4 ± 0.9*
CK (U l^{-1})	115 ± 54	175 ± 95	144 ± 57
LDH (U l^{-1})	193 ± 36	225 ± 41	190 ± 25
IL-1β (pg ml^{-1})	0.16 ± 0.02	0.16 ± 0.01	0.16 ± 0.02
IL-6 (pg ml^{-1})	0.39 ± 0.23	0.45 ± 0.24	0.51 ± 0.28
IL-10 (pg ml^{-1})	0.93 ± 0.07	0.84 ± 0.10	1.05 ± 0.18†
TNFα (pg ml^{-1})	1.30 ± 0.4	0.81 ± 0.2	1.40 ± 0.9

Data are expressed as mean ± SD

DOMS delayed onset muscle soreness, *CK* creatine kinase, *LDH* lactate dehydrogenase, *IL-1b* interleukin-1 beta, *IL-6* interleukin-6, *IL-10* interleukin-10, *TNF-α* tumor necrosis factor-α

*Significantly different from CON and ACUTE ($P < 0.05$). †Significantly different from ACUTE ($P < 0.05$)

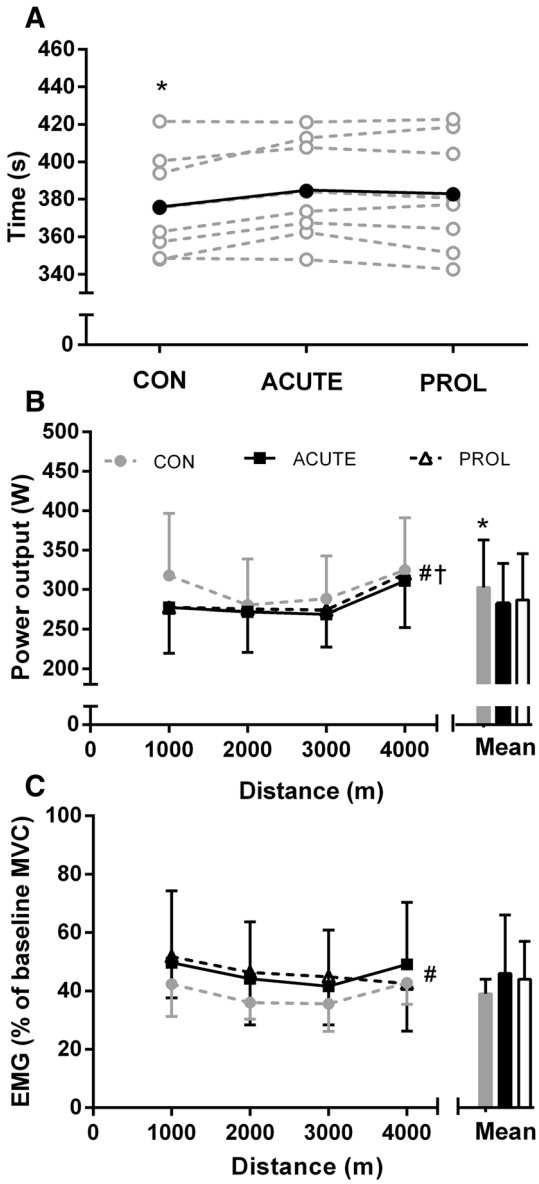

Fig. 4 Individual endurance performance, power output distribution and electromyographic activity during a 4-km cycling time trial immediately after 100-drop jumps (ACUTE), 48 h after 100-drop jumps (PROL), and without prior drop-jumps (CON). Data are expressed as an individual (open circle) and mean (black circle) values. *Statistically lower than ACUTE and PROL ($P < 0.05$). #Significant main distance effect ($P < 0.05$). †Significant main condition effect ($P < 0.05$)

and $Q_{tw.pot}$ were significantly ($P < 0.05$) reduced from baseline after all time trials (Table 1). There was a greater reduction ($P < 0.01$) in $Q_{tw.pot}$ in ACUTE compared with the PROL and CON (Fig. 5). Interestingly, when observed from pre-TT to post-TT, there was no significant decrease in the neuromuscular function in ACUTE (Table 1). The M-wave amplitude was significantly increased ($P < 0.05$) after the time trial in CON and PROL (Table 1). This increase in M-wave amplitude following 4-km TT in CON was significantly different ($P = 0.01$) when compared to $ACUTE_{4\text{-km TT}}$ (Fig. 5). The VA after time trials did not differ between conditions (Fig. 5).

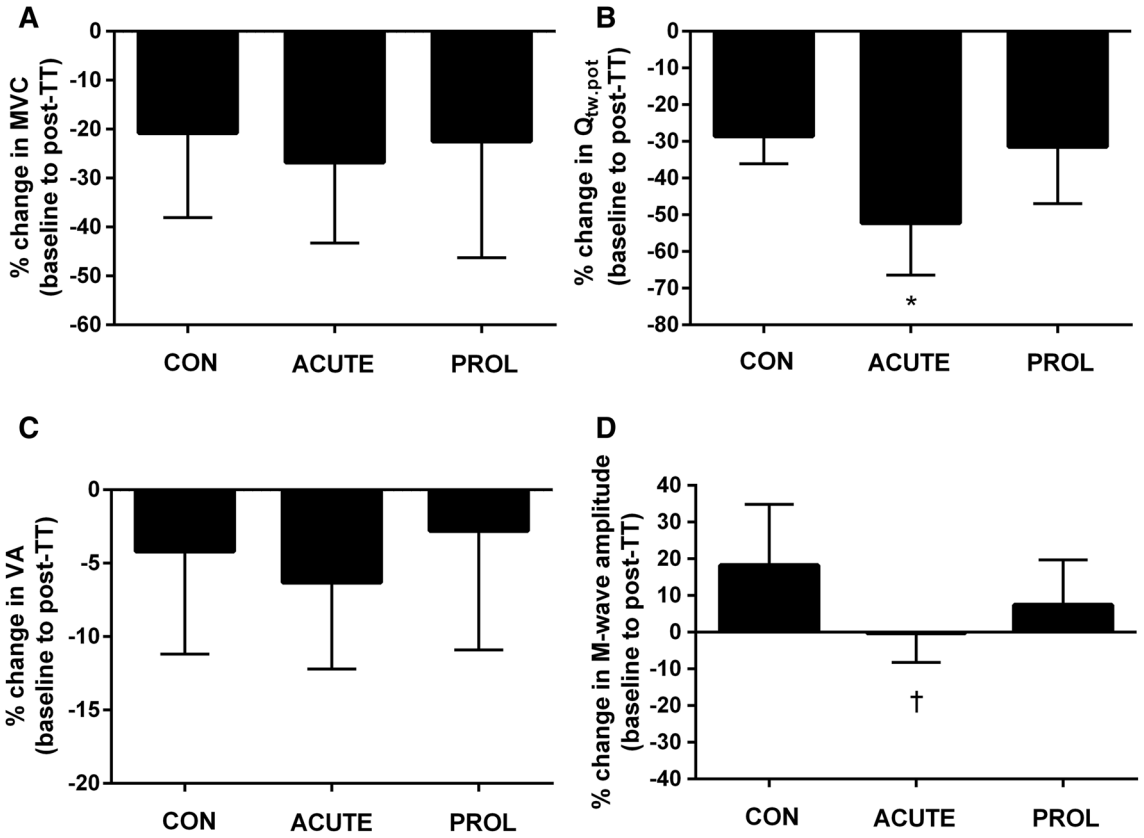

Fig. 5 Percentage of change in neuromuscular function from baseline to post high-intensity endurance exercise. *MVC* maximal voluntary contraction, $Q_{tw.pot}$ potentiated quadriceps twitch-force, *VA* voluntary activation, *M-wave* M-waves amplitude. In the PROL condition, alterations in neuromuscular function were expressed as a percentage of changes from baseline of the ACUTE condition. Data are expressed as mean ± SD. *Significantly different from CON and PROL ($P < 0.05$). †Significantly different from CON ($P < 0.05$)

Discussion

While previous studies have examined the effects of SSC on endurance performance, a novel aspect of the present study was investigating the effect of SSC on the central and peripheral fatigue after endurance performance. The main findings of this study were: (1) previous SSC protocol produced a considerable level of central and peripheral fatigue before the 4-km TT, which was accompanied by an impaired 4-km TT performance in ACUTE, (2) peripheral fatigue level after the 4-km TT was greater in ACUTE when compared with CON and PROL conditions, (3) the impairment in 4-km TT performance 48 h after the SSC protocol was accompanied by an increased DOMS, and (4) changes in central and peripheral components of neuromuscular fatigue in PROL were similar to those observed in CON before and after 4-km TT.

The present study demonstrated that the percentage of change in the knee extensor strength from baseline to pre-TT was reduced more in ACUTE (−25%) than CON (−4%), but without a significant difference when compared to PROL (−10%) or between CON and PROL. Our results also revealed that reduction in the MVC immediately after SSC protocol was accompanied by both peripheral ($Q_{tw.pot}$ −49.2%) and central (VA −6.5%) fatigue, whereas similar responses were not found 48 h after SSC protocol ($Q_{tw.pot}$ −4.4%, and VA −0.9%). These findings show that the acute impact of the SSC on the ability of skeletal muscle to produce force is determined by peripheral and central factors and that a full recovery was achieved 48 h after SSC exercise. These results are, at least in part, in agreement with findings of previous studies (Nielsen et al. 2005; Skurvydas et al. 2000, 2011; Miyama and Nosaka 2007). Significant reduction in both potentiated twitch torque and voluntary activation immediately after exercises to induce muscle damage is often observed (Behrens et al. 2012; Prasartwuth et al. 2005; Doguet et al. 2016; Racinais et al. 2008; Skurvydas et al. 2000, 2011). In contrast, these studies have found different time-course of neuromuscular fatigue recovery in the days following exercises to induce muscle damage. Specifically, studies have demonstrated a full recovery of central fatigue within 24 h (Behrens et al. 2012; Doguet et al. 2016), 48 h (Prasartwuth et al. 2005) and 72 h (Racinais et al. 2008) after eccentric exercise. In respect to peripheral fatigue, it

has been evidenced a full recovery in 24 h (Racinais et al. 2008), 48 h (Behrens et al. 2012) and > 96 h (Prasartwuth et al. 2005; Doguet et al. 2016; Skurvydas et al. 2011). The likely explanation for these different findings may be the different protocol used to induce muscle damage and asses the central and peripheral fatigue. Although the mechanisms involved in the reduction of neural drive immediately after SSC are not fully known, some possibilities have been proposed, such as cortical neurotransmitter depletion, presynaptic or postsynaptic inhibitory effects coming from different feedback sources, disfacilitation of the Ia-afferents coming from the muscle spindles, inhibitory feedback from group III and IV mechanoreceptors and nociceptors (Nicol et al. 2006). Collectively, these findings indicate that the athletes initiated the 4-km TT in ACUTE with exacerbated level of peripheral fatigue; therefore, the negative impact on the contractile function of the skeletal muscle should not be neglected in the previous studies that used the SSC exercise to explore the limiting factors of endurance performance.

The SSC exercise is used to explore the importance of the locomotor muscles for different fatigue theory models in endurance exercise because it produces significant force reduction without altering muscle metabolites (Pi, PCr, ATP, lactate), which are traditionally linked with fatigue accumulation and impaired high-intensity endurance performance (Nielsen et al. 2005). However, it is important to note that no study has assessed the development of central and peripheral fatigue during endurance exercise when preceded by SSC protocol and; therefore, the mechanisms driving to a reduced high-intensity endurance exercise performance immediately after SSC exercise remained unknown. In the present study, the athletes initiated the 4-km TT with an elevated degree of central and peripheral fatigue; however, peripheral fatigue remained elevated after its completion, but without an additional accumulate. This finding is in accordance with a previous observation showing no peripheral quadriceps fatigue accumulation after a 5-km cycling TT preceded by a high-intensity endurance constant-workload exercise performed until exhaustion (Amann and Dempsey 2008). Together, these results suggest that, when a prior fatiguing task promotes a greater level of peripheral fatigue than a subsequent task, an additional accumulation of peripheral fatigue is not observed. In this context, a more complex interplay between central and peripheral sites for the development of neuromuscular fatigue has been recently proposed (Amann et al. 2009). It has been suggested that the degree of peripheral fatigue is limited by a reduction in the central motor drive via inhibitory feedbacks from group III/IV muscle afferents, in order to not exceed a certain level of individual critical threshold of peripheral fatigue at the end of exercise (Amann et al. 2009). Although our study does not allow us to draw a definitive conclusion concerning the mechanisms involved in the adjustment of the power output, it is plausible to assume that the impaired 4-km TT performance in the ACUTE was mediated by both pre-exercise peripheral and central fatigue.

Interestingly, despite the neuromuscular function had been fully recovered 48 h after SSC protocol, there was a significant impairment in 4-km TT performance in PROL compared to CON. This is in accordance with previous studies showing a reduction (4–6%) in high-intensity endurance exercise performance 48 h after SSC exercise (Marcora and Bosio 2007; Burt and Twist 2011). This deleterious effects of the previous SSC have been attributed to increased muscle soreness and concentration of blood inflammatory cytokine, which in theory would signal to the central nervous system and contribute to the development of central fatigue (Marcora and Bosio 2007). However, to the best of our knowledge, the present study was the first to assess a possible linkage between inflammatory responses and neuromuscular fatigue induced by 4-km TT performed 48 h after the SSC protocol. Although the central and peripheral components of neuromuscular fatigue were not different from CON, IL-10 and DOMS were altered 48 h after SSC protocol. The IL-10 was significantly higher 48 h after SSC protocol than ACUTE, indicating an increased anti-inflammatory response. This higher systemic anti-inflammatory response seems to explain the similar plasma concentrations of pro-inflammatory cytokines for these conditions since IL-10 inhibits the production of IL-1β, IL-6 and TNF-α (Pretolani 1999). Furthermore, an increased IL-10 suggests that a pro-inflammatory response seems to have occurred in the period between the termination of the SSC protocol and 48 h subsequent. Another explanation for this result is that systemic and local inflammatory responses can present different time courses (Peake et al. 2005). DOMS also was increased 48 h after the SSC protocol when compared to CON and ACUTE. Although the mechanisms involved in muscle soreness are not completely elucidated, it has been recently proposed that DOMS would be related more with inflammation in the extracellular matrix, rather than myofiber damage and/or inflammation (Peake and Neubauer 2017). It is in accordance with our results showing an increase in DOMS 48 h after SSC protocol even with full recovery of the muscle capacity to produce force. Taken together, these findings suggest that impairment in 4-km TT performance days after SSC protocol may occur mostly due to greater DOMS response, which would be mediated by inflammatory responses rather than central or peripheral fatigue.

Previous studies have suggested that the impairment in endurance performance immediately and days after the exercise to induce muscle damage is due, mainly, to an increased RPE (Marcora et al. 2008; de Morree and Marcora 2013; Marcora and Bosio 2007). It has been proposed that power output distribution (i.e. pacing strategy) could be regulated in a way that RPE does not reach its maximum before the end of the TT (Joseph et al. 2008). Our

results provide further support for this hypothesis; while our participants presented similar RPE during 4-km TT between conditions, the mean power output was lower in ACUTE and PROL compared to CON. However, the novel findings of the current study demonstrate that RPE seems to be affected by different factors in ACUTE and PROL conditions. In fact, in the ACUTE was found a high degree of central and peripheral fatigue, whereas in the PROL a high DOMS was observed. These results suggest that RPE responses during TT were mediated by neuromuscular alterations and muscle soreness immediately and 48 h after intense SSC protocol, respectively.

The CK and LDH activity increased immediately and 48 h after 100-DJ, but not sufficient to reach the level of significance ($P \leq 0.05$). A possible explanation for this result is that trained endurance individuals may present a peak in the CK and LDH activity within 24 h after intense exercise, returning to baseline levels in 48 h (Evans et al. 1986). This seems to be in line with the full recovery of the muscular function at 48 h, suggesting that intense SSC protocol used in the present study may not have been able to induce any muscle damage 48 h after 100-DJ since the loss of strength is considered the best indicator of exercise-induced muscle damage. Another interesting result found in the present study was reduced M-wave amplitude immediately after the SSC protocol and increased M-wave amplitude after 4-km TT in all conditions. A decrease in M-wave amplitude has often been interpreted as membrane excitability impairment; however, this idea has recently been challenged. Indeed, Rodriguez-Falces and Place (2018) have proposed that a decrease in membrane excitability is reflected by an increase rather than a decrease of M-wave amplitude.

It is necessary to acknowledge some of the limitations of the present study. First, is not known to what extent does reductions in isometric muscle function translate to impairment in dynamic exercise performance. Second, the unavoidable time delay (i.e., 2 min) between the end of 4-km cycling TT and the start of neuromuscular testing likely allowed for some recovery, resulting in an underestimation of the real degree of both exercise-induced central and peripheral fatigue. Third, the single electrical stimulation used in our study may have underestimated the level of central fatigue since double-pulse stimulation is considered to be a better methodology. Although some degree of recovery might have existed, transition timing was maintained constant and, therefore, any possible effect of recovery was similar between conditions. Likewise, the use of the single electrical stimulation probably promoted similar underestimation of the level of central fatigue in all conditions. Finally, trained individuals may be less susceptible to the effects of the SSC exercise in the skeletal muscle and, therefore, caution should be exercised when extrapolating these findings to other populations.

Conclusions

In summary, the results of the current study showed that endurance performance in a high-intensity cycling exercise is differently impaired when performed immediately and 48 after intense SSC protocol. The acute impairment in endurance exercise performance was accompanied by a modulation in the central and peripheral sites of the neuromuscular fatigue, whereas prolonged impairment had a perceptual (i.e., DOMS) rather than neuromuscular (i.e., central and peripheral mechanisms) origin. These findings could help exercise physiologists to elaborate more precise strategies with the objective of minimizing the modifications in neuromuscular function (e.g. caffeine) and sensation of pain (e.g. acetaminophen) caused by intense SSC exercise.

Acknowledgements The authors thank the athletes for their participation in this study. Marcos D. Silva-Cavalcante and Patricia G. Couto are grateful to Coordination of Improvement of Personnel of Superior Level (CAPES) for PhD scholarships. Marcos D. Silva-Cavalcante also is grateful to CAPES for current Post Doctoral Fellowship (PNPD/CAPES). The English text of this paper has been revised by Sidney Pratt, Canadian, MAT (The Johns Hopkins University), RSAdip-TESL (Cambridge University).

Author contributions Formulation of the idea and designed research by MDSC, RB and AELS; data collection by MDSC, PGC and RAA; data analysis and interpretation by MDSC, RB, AFG, AELS, PGC and RAA; preparation of the manuscript by MDSC, RB, AFG, AELS, PGC and RAA; edition and revision by MDSC, RB, AFG, AELS, PGC and RAA. All authors have read and give final approval of this version of the manuscript for publication.

Compliance with ethical standards

Conflict of interest The authors declare no conflict of interest.

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