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# Recovery from fatigue following cycling time-trials in elite endurance athletes

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**Ducrocq GP, Hureau TJ, Meste O and Blain.** *Recovery from fatigue following cycling time-trials in elite endurance athletes.*

**Introduction:** We determined the recovery from neuromuscular fatigue in six professional (PRO) and seven moderately trained cyclists (MOD) following repeated cycling time-trials of various durations.

**Method:** Participants performed two 1-min ( $1_{\min}TT$ ) or two 10-min ( $10_{\min}TT$ ) self-paced cycling time-trials with 5-min of recovery in-between. Central and peripheral fatigue were quantified via pre- to post-exercise (15-s through 15-min recovery) changes in voluntary activation (VA) and potentiated twitch force. VA was measured using the interpolated twitch technique and potentiated twitch force was evoked by single ( $QT_{\text{single}}$ ) and paired [10-Hz ( $QT_{10}$ ) and 100-Hz ( $QT_{100}$ )] electrical stimulations of the femoral nerve.

**Results:** Mean power output was 32-72% higher during all the time-trials and decreased less (-10% vs -13%) from the first to second time-trial in PRO compared with MOD ( $P < 0.05$ ). Conversely, exercise-induced reduction in  $QT_{\text{single}}$  and  $QT_{10}/QT_{100}$  was significantly lower in PRO following every time-trial ( $P < 0.05$ ). Recovery from fatigue from 15-s to 2-min for  $QT_{\text{single}}$  and  $QT_{10}/QT_{100}$  was slower in PRO following every time-trial ( $P < 0.05$ ). In both groups, the reduction in  $QT_{\text{single}}$  was lower following the  $10_{\min}TT$ s compared with  $1_{\min}TT$ s ( $P < 0.05$ ). Conversely, VA decreased more after the  $10_{\min}TT$ s compared with  $1_{\min}TT$ s ( $P < 0.05$ ).

**Conclusion:** Our findings showed that excitation-contraction coupling was preserved following exercise in PRO compared with MOD. This likely contributed to the improved performance during repeated cycling time-trials of various durations in PRO, despite a slower rate of recovery in its early phase. Finally, the time-course of recovery from neuromuscular fatigue in PRO was dependent on the effects of prolonged low-frequency force depression.

**Key words:** *neuromuscular fatigue, recovery, muscle activation, exercise performance, cycling time-trial, professional cyclists*

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## INTRODUCTION

A relatively large number of findings on the metabolic, cardiovascular and respiratory bases of exercise performance in elite endurance athletes has accumulated over the last century (see 1 for review), although much remains to be learned about the mechanisms that contribute to neuromuscular fatigue development and recovery during physical exercise in these athletes.

Exercise-induced neuromuscular fatigue is a reversible impairment of the muscle's ability to produce

force or power (2). The mechanisms underpinning neuromuscular fatigue involves the impairment of the muscle's ability to produce force in response to neural input (i.e. peripheral fatigue, 3) and/or the failure (or compromised willingness) of the central nervous system to fully activate the exercising muscle (i.e. central fatigue, 2). Specifically, the disruption of intramuscular homeostasis from the accumulation of metabolic by-products such as hydrogen ions and inorganic phosphate determines peripheral fatigue by compromising the excitation-contraction coupling of myocytes (3-5). Decreased motoneuronal excitability (2,6), depletion of brain neurotransmitters (7) and/or inhibitory inputs from metabo- and mecanosensitive muscle afferent fibers (8,9) contribute to central fatigue. These processes compromise physical exercise tolerance/performance in health and disease, in particular at intensities that provoke the accumulation of the intramuscular metabolites associated with peripheral fatigue (i.e. above the critical velocity / power, 10,11). Neuromuscular fatigue is therefore a key determinant of endurance performance (8,12-15).

Comprehensive findings on the neuromuscular consequences of severe-intensity exercise in elite/professional endurance athletes can provide important insights about the physiological limits of human performance. The reduction in the ATP cost of contraction and in the accumulation of the intramuscular metabolites associated with peripheral fatigue (e.g., inorganic phosphate, hydrogen ions, etc.) during exercise in endurance-trained athletes might prevent severe exercise-induced neuromuscular fatigue (16,17). To our knowledge, only one study (18) has attempted to characterize the neuromuscular consequences of exhaustive exercise in highly trained endurance athletes [i.e. maximal oxygen consumption ( $\dot{V}O_{2\text{ MAX}}$ ) averaging 74 mL.min<sup>-1</sup>.kg<sup>-1</sup>]. Findings showed that, despite higher exercise performance, peripheral fatigue was lower following repeated intermittent isometric contractions in the highly trained athletes compared with age-matched untrained participants. There is, however, uncertainty about the degree to which findings from single-limb exercise apply to endurance exercise such as cycling or running, both of which require greater muscle mass and have higher metabolic, cardiovascular and ventilatory requirements (19-21). Findings to characterize the contributions of peripheral and central mechanisms to fatigue and the time-course of recovery following whole-body exercise are thus needed to accurately determine the

neuromuscular consequences of severe-intensity exercise in elite/professional endurance athletes.

It is also not clear from the very limited existing literature whether exercise intensity and duration in the severe-intensity domain influences the etiology and the degree of exercise-induced neuromuscular fatigue. For example, a greater impairment in the potentiated twitch force was found following a ~3-min compared with an ~11-min constant workload cycling test to exhaustion (22). In contrast, no significant difference between end-exercise reductions in the potentiated twitch force was observed when participants performed a 3 or 12-min severe intensity cycling exercise that led to the depletion of the total amount of work that can be performed above the critical power (23). Moreover, the end-exercise changes in metabolic variables associated with muscle fatigue, such as the concentration of the hydrogen ion ( $H^+$ ), phosphocreatine, glycogen or the potassium ion ( $K^+$ ) were found to be similar when severe intensity exercises of duration ranging from ~2 to ~14-min were performed to exhaustion (10). Further data are therefore needed to elucidate the role of exercise intensity and duration on neuromuscular fatigue during cycling exercise within the severe intensity domain.

Finally, no systematic attempts have been made to characterize the mechanisms that determine the time-course of recovery from neuromuscular fatigue following whole-body exercise or how these mechanisms are modulated by exercise intensity, duration and repetition, especially in elite endurance athletes. Such findings would be of significant practical use to enhance exercise prescription and improve exercise performance.

To address the lack of neuromuscular fatigue data on elite endurance athletes, we compared the neuromuscular function of professional and moderately trained cyclists during and immediately after repeated self-paced cycling time-trials of various durations within the severe-intensity domain. Our goal was to I) accurately characterize the contribution of peripheral and central mechanisms to recovery from neuromuscular fatigue and II) determine how these mechanisms are affected by the intensity/duration and repetition of successive exercise trials to exhaustion.

## METHOD

### *Participants*

Six professional road cyclists (mean  $\pm$  SD; age,  $27 \pm 3$  years; height,  $180 \pm 6$  cm; weight,  $68 \pm 4$  kg; body fat,  $10.3 \pm 1.6\%$ ; maximal oxygen uptake:  $\dot{V}O_{2max}$ ,  $75 \pm 5$  ml.min<sup>-1</sup>.kg<sup>-1</sup>; peak power output,  $6.1 \pm 0.4$  W.kg<sup>-1</sup>) and seven moderately trained cyclists (age,  $22 \pm 2$  years; height,  $181 \pm 6$  cm; weight,  $73 \pm 13$  kg; body fat,  $13.6 \pm 4.4\%$ ;  $\dot{V}O_{2max}$ ,  $53 \pm 8$  ml.min<sup>-1</sup>.kg<sup>-1</sup>; peak power output,  $3.9 \pm 0.5$  W.kg<sup>-1</sup>) participated in this study. Professional cyclists were competing at the continental pro level and trained on average  $20 \pm 7$  h per week. Moderately trained

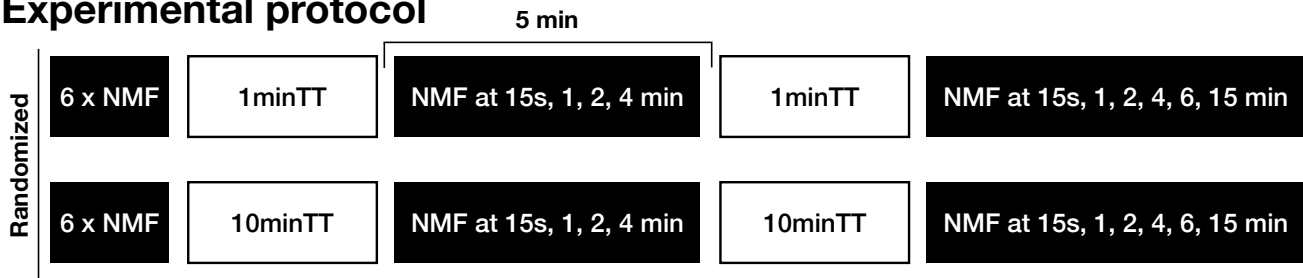
cyclists were competing at a local level and trained on average  $6 \pm 2$  h per week. All participants were non-smokers, not medicated, free of orthopedic injury of the lower limb and asymptomatic for cardiovascular, respiratory or neuromuscular disorders. Participants were asked to avoid physical exercise and caffeine consumption 24h before each visit. Written informed consent was obtained from each subject prior to the beginning of the study. The study was approved by the local ethics committee and conducted according to the Declaration of Helsinki for human experimentation.

### *Experimental design*

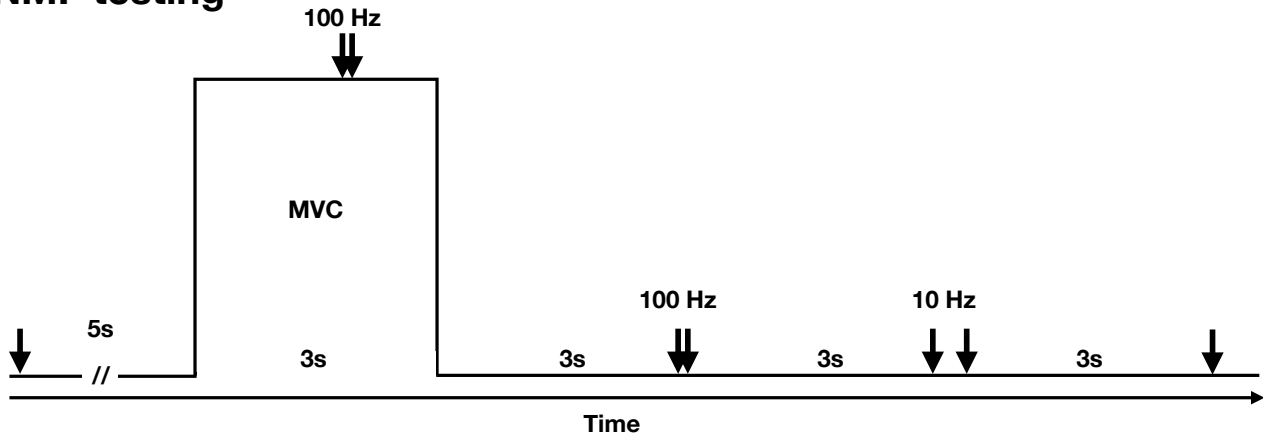
During preliminary visits, anthropometric measurements were collected and participants were thoroughly familiarized with the neuromuscular and exercise testing procedures. These preliminary visits included at least four practice sessions during which participants performed, on separated days, two 1-min and two 10-min time-trials on a stationary cycle ergometer (Wattbike, Nottingham, UK), with freedom to alter the power output by changing the resistance and/or the pedaling frequency (i.e. self-paced). The time elapsed during the time-trial was the only information displayed on a monitor placed directly in front of them. They were given strong vocal encouragement and were instructed to remain seated throughout the exercise. Familiarization with the time-trials was considered accomplished when the difference in mean power output between two practice trials of the same exercise duration was lower than 2 % (24). Otherwise, participants were asked to perform additional practice sessions until this criterion was met.

During the first experimental session, participants performed a maximal and graded exercise test (professional cyclists: 80W + 40W every two minutes; moderately trained cyclists: 70W + 35W every two minutes) to determine the peak power output and  $\dot{V}O_{2max}$ . To ensure that participants reached  $\dot{V}O_{2max}$  during the maximal and graded exercise test, participants were asked to perform, after a ten minute recovery period, a constant workload trial until exhaustion at a power output corresponding to 105% of the peak power output reached during the maximal and graded exercise test (25). In all participants, the peak  $\dot{V}O_2$  from the constant workload trial was not different ( $P = 0.80$ ) than the peak  $\dot{V}O_2$  from the maximal and graded exercise test ( $62.6 \pm 14.5$  ml.min<sup>-1</sup>.kg<sup>-1</sup> vs  $62.3 \pm 15.0$  ml.min<sup>-1</sup>.kg<sup>-1</sup>, respectively), indicating that every participant reached  $\dot{V}O_{2max}$  during the maximal and graded exercise test. During the subsequent experimental visits (Figure 1), participants performed on separate days a self-paced 1-min time-trial and a self-paced 10-min time-trial. To determine the effect of exercise repetition on recovery from neuromuscular fatigue, they performed a second time-

## Experimental protocol



## NMF testing



**Figure 1. Timeline of the experimental protocol and of the standardized set of contractions used to assess neuromuscular fatigue.**

Upper panel illustrates the experimental sessions involving the assessment of the neuromuscular function (NMF) prior and after two 1-min (1minTT) and two 10-min (10minTT) cycling time-trials. Lower panel illustrates the standardized set of contractions used to assess the neuromuscular function prior and after exercise (i.e. NMF in the upper panel). MVC: maximal voluntary contraction; black arrow: supramaximal electrical stimulation of the femoral nerve.

trial of similar duration [i.e., 1-min or 10-min], five minutes after the end of the first time-trial. Importantly, to ensure that performance during the first time-trial was maximal, participants were first informed that they would perform only one time-trial. After performing the first-time trial, they were then asked to perform the second one. Neuromuscular testing (detailed below) was performed prior to exercise and immediately after the first and second time-trials. Experimental sessions were conducted at the same time of day and separated by 2-6 days. To account for potential order effects, a randomized block design was used for the two experimental visits involving the cycling time-trials.

### Data collection and analysis

#### Neuromuscular function

*Contractile function and voluntary activation of the quadriceps.* For the assessment of contractile function, subjects were seated on a custom-made bench, arms folded across the chest, with a trunk/thigh angle of 135° and the right knee joint angle at 90°. A non-compliant strap attached to a calibrated load cell (model SM-2000N, Interface, Scottsdale, AZ, USA) was fixed to the participant's right ankle, just superior to the malleoli. The

cathode, a self-adhesive electrode (3 x 3 cm, Ag-AgCl, Mini-KR, Contrôle-Graphique, Brie-Comte-Robert, France), was placed on the femoral triangle, at the stimulation site that resulted in both maximal force output and maximal amplitude of the compound muscle action potential ( $M_{MAX}$ ) for the *vastus lateralis* (VL), *vastus medialis* (VM) and *rectus femoris* (RF). The anode, a carbon-impregnated electrode (70 x 50 mm), was rubbed with conductive gel and placed on the gluteal fold. The position of these electrodes was marked with indelible ink to ensure a reproducible stimulation site across visits. A constant current stimulator (DS7A, Digitimer, Hertfordshire, UK) delivered a square wave stimulus (1 ms) at a maximum of 400 V. To assure maximal spatial recruitment of motor units during the neuromuscular tests, the stimulation intensity ( $155 \pm 52$  mA) was set to 120% of the stimulation intensity eliciting maximal quadriceps twitch and M-wave peak-to-peak amplitude with increasing stimulus intensities. No electrical activity of the *biceps femoris* (BF) was observed during stimulation. Neuromuscular assessment of the quadriceps muscle was conducted prior and immediately after every cycling time-trials using a standardized set of contractions. In each set, an

unpotentiated twitch was evoked 5-s prior a 3-s maximal voluntary contraction (MVC) using a single supramaximal electrical stimulation of the femoral nerve. During MVC, superimposed paired stimuli at 100 Hz ( $QT_{100, \text{superimposed}}$ ) were delivered during the peak force of the MVC to determine voluntary activation of the quadriceps (VA, 26). Then, potentiated quadriceps twitch force evoked by paired [100 Hz ( $QT_{100}$ ) and 10 Hz ( $QT_{10}$ )] and single ( $QT_{\text{single}}$ ) electrical stimuli of the femoral nerve were elicited 3, 6 and 9-s after MVC, respectively. Six sets, separated by 1-min, were performed at baseline (i.e. before the first cycling time-trial). Following exercise, measurements of the neuromuscular function were obtained using the same standardized set of contractions at exactly 15-s and 1-, 2- and 4-min post-exercise, in order to capture the rapid recovery from fatigue that occurs within the first minutes following exercise termination. After the second 1-min and 10-min cycling time-trials, we also performed neuromuscular measurements at 6- and 15-min post-exercise. For all  $QT_{10}$ ,  $QT_{100}$ , and MVCs, we determined peak force. For all  $QT_{\text{single}}$ , we assessed peak force, contraction time to peak force (i.e. the time between stimulus and peak twitch force), maximal rate of force development (i.e. maximal value of the first derivative of the force signal), and half relaxation time (i.e. time between peak twitch force and half of the decline in maximal twitch amplitude). The  $QT_{10}/QT_{100}$  ( $QT_{10:100}$ ) ratio was calculated, as a decrease in this ratio is commonly interpreted as an index of prolonged low-frequency force depression (27). Quadriceps VA was calculated according to the following formula:  $VA (\%) = (1 - QT_{100, \text{superimposed}}/QT_{100}) \times 100$ . Baseline values for MVC peak force, maximal rate of force development, VA,  $QT_{10:100}$ ,  $QT_{\text{single}}$ ,  $QT_{10}$  and  $QT_{100}$  peak force were calculated by averaging the three highest values from the pre-exercise standardized sets of contractions. Baseline values for twitch contraction time and twitch half-relaxation time were calculated by averaging the three lowest values from the pre-exercise standardized sets of contractions. Pre- to post-exercise difference in MVC, VA,  $QT_{\text{single}}$ ,  $QT_{10}$ ,  $QT_{100}$  and  $QT_{10:100}$  (expressed as a percent change from pre-exercise) were calculated to quantify and characterize the origin of exercise-induced neuromuscular fatigue.

To quantify the rate of recovery over time, the time-course of recovery was divided into three periods: I) from 15-s to 2-min, II) from 2-min to 4-min, and III) from 4-min to 15-min. The recovery rate for each period was calculated as follows:  $\text{Recovery (in \% / min)} = 100 \times [(X_{t_1} - X_{t_2})/X_{\text{Fat}}]/T_{\text{recovery}}$ , with  $X_{t_1}$  and  $X_{t_2}$  corresponding to the exercise-induced reduction in the index (in %) measured at the beginning ( $t_1$ ) and end ( $t_2$ ) of the recovery period,  $X_{\text{Fat}}$  corresponding to the highest level of exercise-induced fatigue (i.e. maximal pre- to post- reduction in a given fatigue index; in %), and  $T_{\text{recovery}}$  the duration of the recovery period (1.75, 2, 11-

min for the first, second and third recovery period, respectively).

To indirectly and qualitatively estimate the difference in muscle fiber type distribution, baseline  $QT_{\text{single}}$  peak force, twitch contraction time and maximal rate of force development were compared between professional and moderately trained cyclists (28). We also calculated quadriceps post-activation potentiation ( $QT_{\text{potentiation}}$ ) using the following formula:  $QT_{\text{potentiation}}(\%) = (QT_{\text{single}} - QT_{\text{unpot}})/QT_{\text{unpot}}$ . Indeed, participants with the highest levels of  $QT_{\text{potentiation}}$  have been shown to exhibit the highest percentage of type II muscle fibers (28).

Finally, the peripheral fatigue cost of exercise was calculated as follows:  $QT_{\text{single}}/W$ , where  $QT_{\text{single}}$  represents the change in  $QT_{\text{single}}$  from baseline to 15 s post-exercise (in N) and  $W$  the total work done during the corresponding exercise (in  $\text{kJ}\cdot\text{kg}^{-1}$  of body mass).

#### *Surface electromyography*

Electromyographic (EMG) activity of the VL, VM, RF and BF of the right leg was recorded during MVCs and during cycling time-trials by four pairs of Ag/AgCl surface electrodes (diameter = 10 mm; inter-electrode distance = 20 mm; Mini-KR, Contrôle-Graphique, Briec-Comte-Robert, France) placed on the muscle belly connected to an EMG system (Octal Bio-Amp, ML138, ADInstrument, Sydney, Australia). A reference electrode was placed on the right lateral tibial condyle. The skin was shaved, abraded with emery paper and cleaned with alcohol to reduce skin impedance below 3 k $\Omega$ . The position of the electrodes optimizing M-wave peak-to-peak amplitude was marked with indelible ink to ensure identical placement at subsequent visits. EMG signals were amplified (gain = 20 mV), filtered (bandwidth frequency, 10 - 500 Hz), and recorded (sampling frequency, 4 kHz) using commercially available software (Labchart 7, ADInstruments, Sydney, Australia). Each burst onset and offset of the EMG signal, recorded during the cycling time-trials, was determined offline using a custom-made Matlab algorithm (Matlab 7.12, MathWorks, Natick, MA, USA). The algorithm consists in filtering the EMG signal with a finite input response filter with linear phase (200-700Hz), followed by a rectification (absolute value) and a smoothing operation with a Gaussian function. A threshold crossing is applied to find the onset and offset of the burst that lasted at least 100ms. Each burst was visually inspected and validated. The root mean square during each MVC was calculated over a 0.5-s interval during the plateau phase of the MVC in order to determine the maximal level of muscle activation during the MVC. During exercise, the root mean square of each burst from the VL, VM and RF was calculated, normalized to the root mean square recorded during pre-exercise MVCs ( $VL_{\%MVC}$ ,  $VM_{\%MVC}$  and  $RF_{\%MVC}$ , respectively), and averaged every 5-s.

#### *Exercise response*

Pulmonary ventilation and gas exchange indices were measured breath-by-breath at rest and throughout time-trials using a stationary automatic ergospirometer (MS-CPX, Viasys, San Diego, California, USA). Before each test, gas analyzers were calibrated using a certified gas preparation ( $O_2$ : 16%;  $CO_2$ : 5%) and an accurate volume of ambient air (2 L) was used to adjust the pneumotachograph. Heart rate was calculated from the R-R interval recorded by a heart rate monitor (RS800CX, Polar Electro, Kempele, Finland). Oxygen uptake ( $\dot{V}O_2$ ), carbon dioxide output ( $\dot{V}CO_2$ ),  $\dot{V}CO_2 \cdot \dot{V}O_2^{-1}$ , minute ventilation,  $\dot{V}_E \cdot \dot{V}O_2^{-1}$ ,  $\dot{V}_E \cdot \dot{V}CO_2^{-1}$ , breathing frequency, tidal volume and heart rate measured during every exercise test were averaged every 5-s. Capillary blood lactate samples (5  $\mu$ l) were collected from a fingertip at rest and 3-min post-exercise after the first and the second time-trials during every experimental visit. Samples were analyzed by an electrochemical method (LactatePro, Arkray, Kyoto, Japan) immediately after sampling using a test strip for determination of blood lactate concentration. Rate of perceived exertion was measured every minute using a CR10 Borg's scale (29). Specifically, subjects were asked to rate how hard, heavy, and strenuous was the preceding minute of exercise.

### Statistical analysis

Data presented in the results section are expressed as mean  $\pm$  SD. Normality of every dependent variable and homogeneity of the variance of the distributions (equal variance) were confirmed using the Kolmogorov-Smirnov test and the Levene test, respectively. To protect against the risk of type I error arising from multiple comparisons, a multivariate analysis (MANOVA) was conducted on our dependent variables recorded during exercise (i.e., power output, EMG and cardiometabolic data) or during post-exercise recovery (i.e., neuromuscular fatigue indices). A significant ( $P < 0.001$ ) *condition*  $\times$  *time* effect was found for both the exercise and post-exercise recovery datasets. To determine the effect of time-trial duration (e.g., the first 1-min time-trial vs. the 10-min time-trial) on time-dependent variables recorded during the time-trial and the post-exercise recovery, two-way ANOVAs with repeated measures (*condition*  $\times$  *time*) were performed on power output, EMG, cardiometabolic data and neuromuscular fatigue indices for each group. To determine the effect of exercise repetition (e.g., the first vs. the second 1-min time-trials) on time-dependent variables recorded during exercise and the post-exercise recovery, two-way ANOVAs with repeated measures (*exercise repetition*  $\times$  *time*) were performed on power output, EMG, cardiometabolic data and neuromuscular fatigue indices for each group. When a significant difference was found, multiple comparisons analysis was performed using Tukey's HSD test. Effect size was assessed using partial eta-squared ( $\eta^2$ ). An  $\eta^2$  index for effect size was considered as small when  $\eta^2$  was lower than 0.07,

medium when  $\eta^2$  was comprised between 0.07 and 0.20, and large when  $\eta^2$  was greater than 0.20 (30). To determine differences between professional and moderately trained cyclists, a categorical (*group*) factor was added to the above-mentioned ANOVAs. When a significant *group* effect was found, Student's unpaired t-test with Bonferroni's correction for multiple comparisons was used for *post-hoc* analysis between groups (18). Effect size was assessed using Cohen's *d* index. A Cohen's *d* index for effect size was considered as small when *d* was lower than 0.35, medium when *d* was comprised between 0.35 and 0.65, and large when *d* was greater than 0.65 (30). From the data measured in the first 1-min and 10-min cycling time-trials, a correlation analysis using the Pearson's correlation coefficient was performed to determine the relationships between reduction in  $QT_{single}$  and mean  $VL_{\%MVC}$ ,  $VM_{\%MVC}$  and  $RF_{\%MVC}$  during exercise, and between the reduction in  $QT_{single}$  and baseline  $QT_{single}$  potentiation. Statistical analyses were conducted using Statistica 8.0 (StatSoft, Inc., Tulsa, OK, USA). Statistical significance was set at  $P < 0.05$ .

## RESULTS

### *Muscle activation and exercise performance*

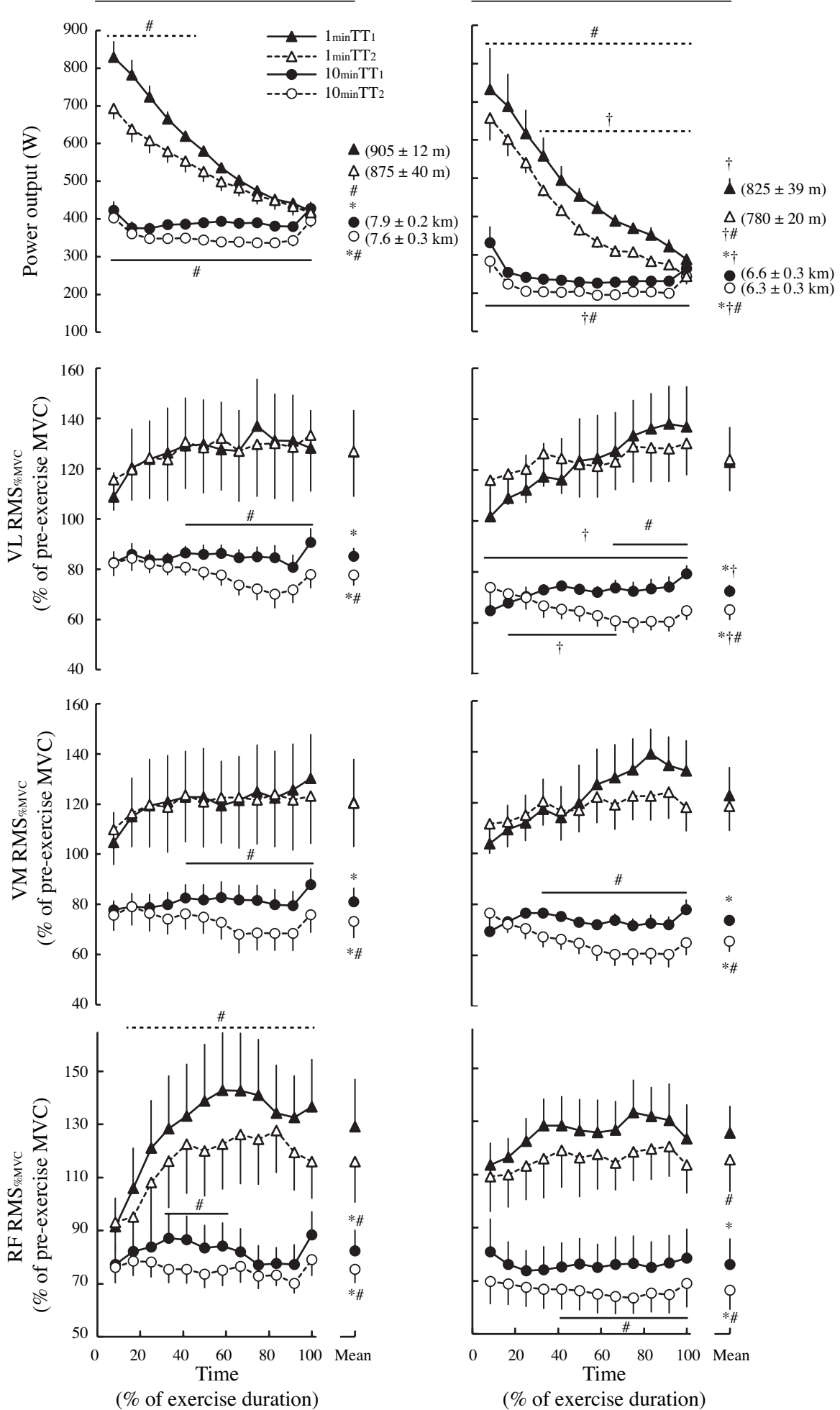
Mean power output ( $P < 0.01$ ,  $d > 2.3$ ) and total distance ( $P < 0.01$ ,  $d > 2.8$ ) in all time-trials were, respectively,  $>24\%$  and  $>10\%$  higher in professional compared with moderately trained cyclists (Fig. 2). For both groups, regardless of exercise duration, mean power output ( $P < 0.05$ ,  $\eta^2 > 0.52$ ) and total distance ( $P < 0.05$ ,  $\eta^2 > 0.55$ ) were significantly reduced from the first to the second time-trial, but these reductions were lower in professional compared with moderately trained cyclists ( $P < 0.01$ ,  $d > 2.9$ ).

Regarding EMG activity, no significant exercise-induced alteration in M-wave peak-to-peak amplitude was found for VL, VM or RF in either group, indicating no alteration in membrane excitability following the 1- and 10-min time-trials (Table 1). During exercise, mean  $VL_{\%MVC}$ ,  $VM_{\%MVC}$  and  $RF_{\%MVC}$  were greater ( $P < 0.05$ ,  $\eta^2 > 0.51$ ) during the 1-min compared with 10-min time-trials in both groups (Fig. 2). In addition, the reduction in power output during the second compared with the first time-trials was associated with a significant reduction in mean  $RF_{\%MVC}$  ( $P < 0.05$ ,  $\eta^2 > 0.32$ ) for the 1-min time-trials and in mean  $VL_{\%MVC}$  ( $P < 0.05$ ,  $\eta^2 > 0.55$ ), mean  $VM_{\%MVC}$  ( $P < 0.05$ ,  $\eta^2 > 0.56$ ) and mean  $RF_{\%MVC}$  ( $P < 0.05$ ,  $\eta^2 > 0.32$ ) for the 10-min time-trials. Mean  $VL_{\%MVC}$  ( $>18\%$ ,  $P < 0.01$ ,  $d = 2.0$ ) were higher in professional compared with moderately trained cyclists during the 10-min time-trials (Fig. 2).

### *Neuromuscular fatigue indices*

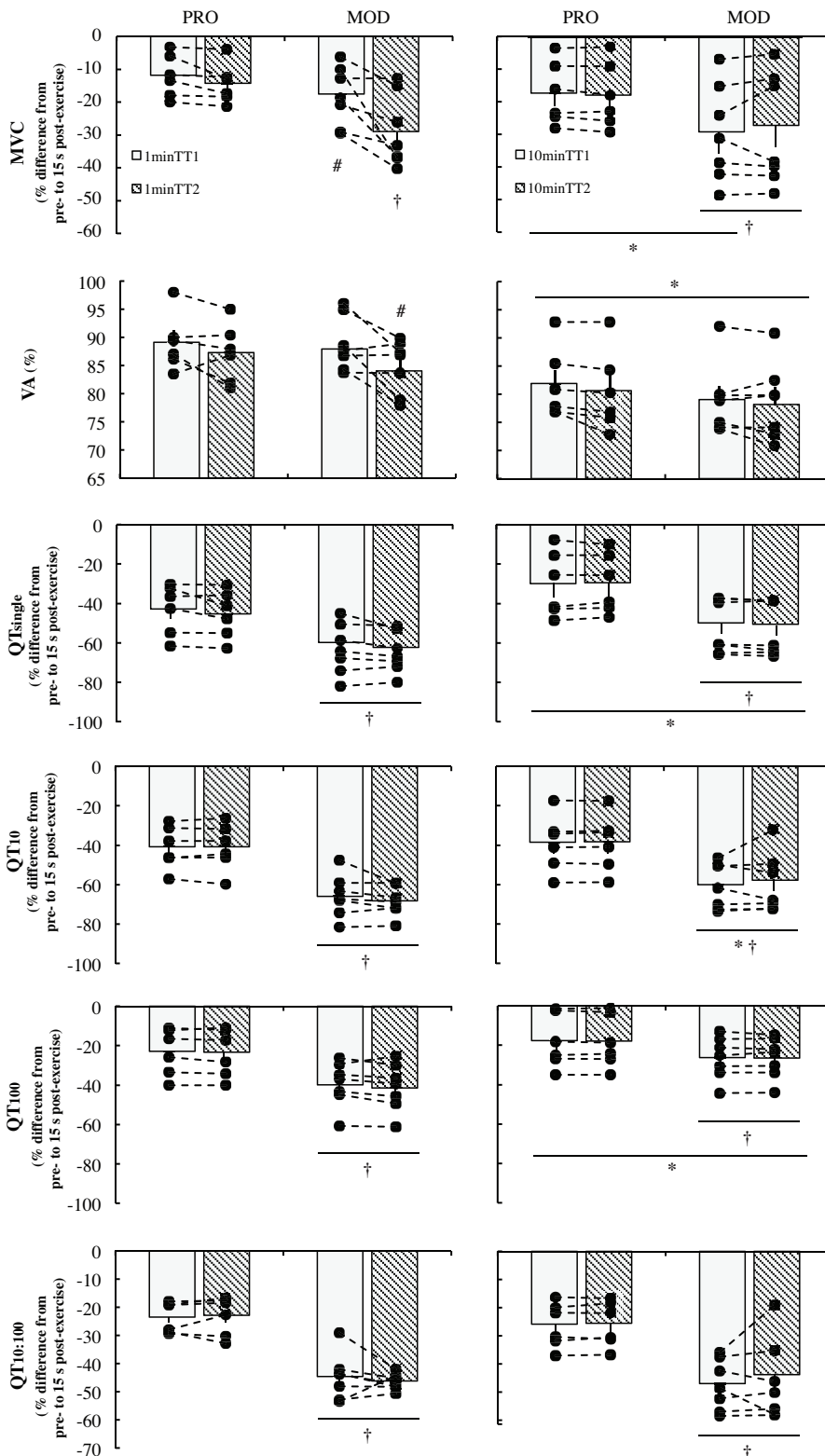
Neuromuscular fatigue indices are presented in Figures 3, 4 and Table 1. At baseline, higher values in twitch contraction time ( $P < 0.01$ ,  $d > 1.51$ ) and twitch half-relaxation time ( $P < 0.05$ ,  $d > 1.55$ ) as well as lower

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PRO MOD



**Figure 2 (previous page). Mean power output, total distance and muscle activation during 1-min and 10-min cycling time-trials.**

Graphical data are presented in the figure as mean  $\pm$  SE. Total distance for each time-trial is presented as mean  $\pm$  SD for both groups into brackets on the upper panels. PRO, professional cyclists; MOD, moderately trained cyclists; VL, *vastus lateralis*; VM, *vastus medialis*; RF, *rectus femoris*; 10<sub>min</sub>TT, 10-min time-trial; 1<sub>min</sub>TT, 1-min time-trial. \*  $P < 0.05$  10<sub>min</sub>TT1 vs. 1<sub>min</sub>TT1 or 10<sub>min</sub>TT2 vs. 1<sub>min</sub>TT2; #  $P < 0.05$  between the first and the second time-trial; †  $P < 0.05$  between PRO and MOD. The horizontal dotted lines encompass time-points for which significant statistical differences between the first and second time-trial or between groups were found for the 1-min time-trials. The horizontal solid lines encompass time-points for which significant statistical differences between the first and second time-trial or between groups were found for the 10-min time-



**Figure 3. Quadriceps neuromuscular fatigue after 1-min and 10-min time-trials.**

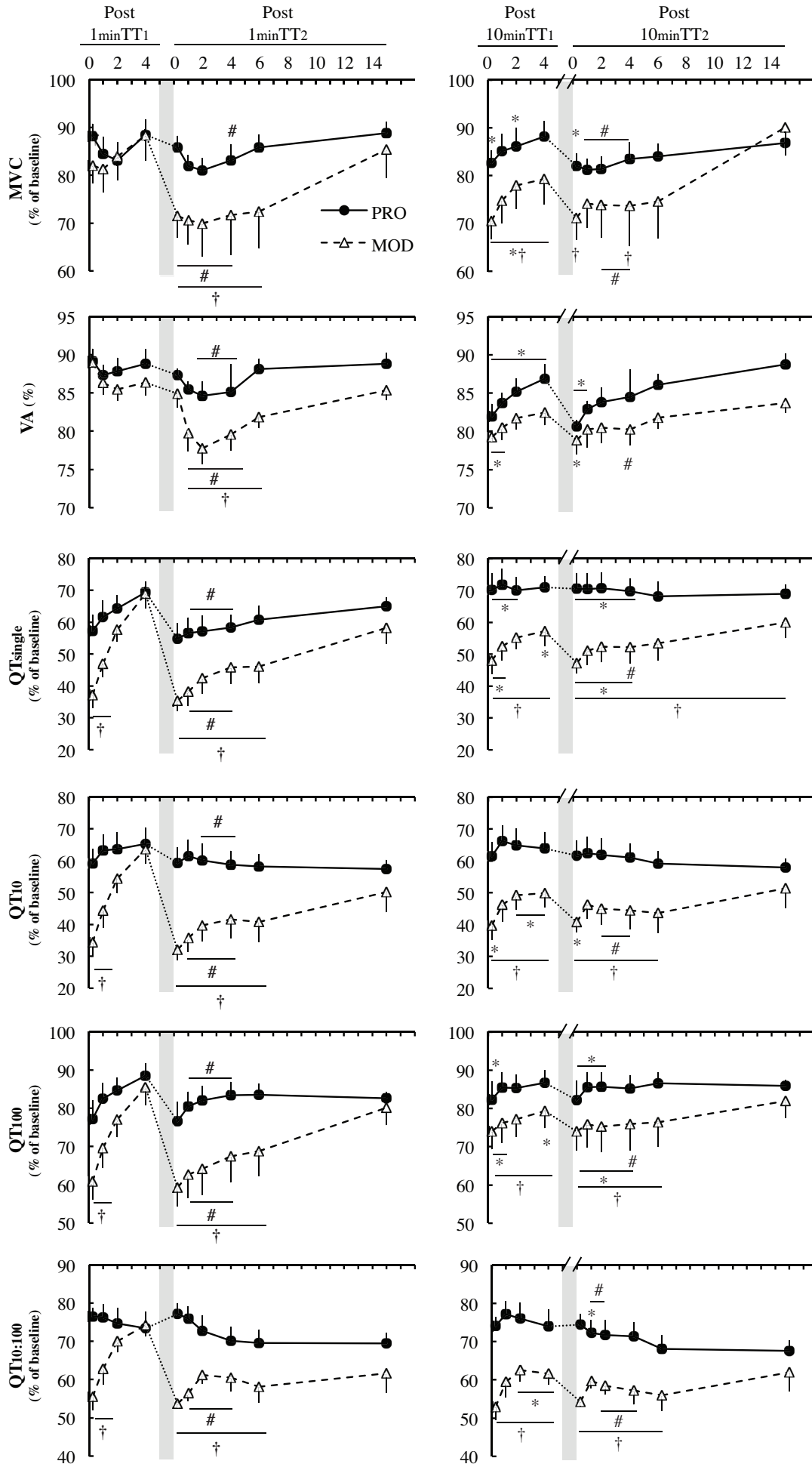
For every index, bars correspond to mean group data and dots to individual data. Data from the 1<sub>min</sub>TTs are presented in the left figures, whereas data from 10<sub>min</sub>TTs are presented in the right figures. PRO, professional cyclists; MOD, moderately trained cyclists; MVC, maximal voluntary contraction; VA, voluntary activation of the quadriceps; QT<sub>single</sub>, potentiated quadriceps twitch evoked with supramaximal electrical single stimulation; QT<sub>10</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 10-Hz paired stimulation; QT<sub>100</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 100-Hz paired stimulation; 10<sub>min</sub>TT, 10-min time-trial; 1<sub>min</sub>TT, 1-min time-trial. \*  $P < 0.05$  between 1-min and 10-min time-trial; #  $P < 0.05$  between first and second time-trial; †  $P < 0.05$  between PRO and MOD. The horizontal line encompasses time-points for which significant statistical differences between the first and second time trial or between groups were found.



# FATIGUE AND RECOVERY IN PROFESSIONAL CYCLISTS

Recovery Time (min)

Recovery Time (min)



**Figure 4 (previous page). Neuromuscular fatigue recovery after 1-min and 10-min cycling time-trials.**

Data are presented as mean  $\pm$  SE. The gray area corresponds to the exercise period of the second cycling time-trial. All neuromuscular fatigue indices remained significantly different from baseline throughout the recovery period. PRO, professional cyclists; MOD, moderately trained cyclists; QT<sub>single</sub>, potentiated quadriceps twitch evoked with supramaximal electrical single stimulation; QT<sub>10</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 10-Hz paired stimulation; QT<sub>100</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 100-Hz paired stimulation; 10<sub>min</sub> TT, 10-min time-trial; 1<sub>min</sub> TT, 1-min time-trial. \*  $P < 0.05$  between 1-min and 10-min time-trial; #  $P < 0.05$  between first and second time-trial; †  $P < 0.05$  between PRO and MOD. The horizontal line encompasses time-points for which significant differences between the first and second time-trial or between groups were found.

values in MVC peak force ( $P < 0.05$ ,  $d > 0.89$ ), QT<sub>single</sub> ( $P < 0.05$ ,  $d > 2.19$ ), QT<sub>10</sub> ( $P < 0.05$ ,  $d > 1.38$ ), QT<sub>100</sub> ( $P < 0.01$ ,  $d > 1.53$ ), QT<sub>potentiation</sub> ( $30 \pm 11$  % vs.  $56 \pm 19$  %,  $P < 0.05$ ,  $d = 1.7$ ) and maximal rate of force development ( $P < 0.05$ ,  $d = 2.53$ ) were found in professional compared with moderately trained cyclists (Table 1). At 15 s post-exercise, all indices were significantly reduced from pre-exercise in both groups ( $P < 0.001$ ,  $\eta^2 > 0.38$ ). The pre- to 15-s post-exercise difference in QT<sub>single</sub> was negatively correlated with baseline QT<sub>potentiation</sub> in both the 1-min and 10-min time-trials ( $R^2 > 0.60$ ,  $P < 0.01$ ; see SDC 1) [This paper cites and discusses four (4) supplemental items - two (2) figures and two (2) tables. Suppl. figure 1 is SDC 1, suppl. table 1 is SDC 2, suppl. table 2 is SDC 3, and suppl. figure 2 is SDC 4]. Smaller reductions in QT<sub>single</sub> ( $P < 0.01$ ,  $d > 1.7$ ), QT<sub>10</sub> ( $P < 0.01$ ,  $d > 1.9$ ), QT<sub>100</sub> ( $P < 0.05$ ,  $d > 0.88$ ), QT<sub>10:100</sub> ( $P < 0.001$ ,  $d > 2.8$ ), maximal rate of force development ( $P < 0.001$ ,  $d > 1.96$ ) and MVC peak force ( $P < 0.05$ ,  $d > 0.87$ ) were found in professional compared with moderately trained cyclists regardless of exercise duration (Fig. 3, Table 1). When normalized by the total amount of work, the peripheral fatigue cost of exercise was significantly lower in professional compared with moderately trained cyclists during every time-trials (Fig. 5).

In both groups, the reductions in QT<sub>single</sub> ( $P < 0.001$ ,  $\eta^2 > 0.69$ ) and QT<sub>100</sub> ( $P < 0.05$ ,  $\eta^2 > 0.31$ ) 15 s after exercise termination were more pronounced after the 1-min compared with 10-min time-trials. The reduction in QT<sub>10</sub> was larger after the 1-min compared to the 10-min time-trials in moderately trained cyclists only ( $P < 0.001$ ,  $\eta^2 = 0.82$ ). The reduction in QT<sub>single</sub> measured after the first 1-min and 10-min time-trials was significantly correlated with the level of VL<sub>%MVC</sub> ( $R^2 = 0.72$ ;  $P < 0.001$ ) and VM<sub>%MVC</sub> ( $R^2 = 0.55$ ;  $P = 0.013$ ) during exercise. When normalized by the total amount of work, the peripheral fatigue cost of exercise was significantly less in the 10-min compared with 1-min time-trials (Fig. 5). Despite significant differences in quadriceps muscle activation and power output during exercise, both groups showed no significant difference in QT<sub>single</sub> ( $P > 0.44$ ), QT<sub>10</sub> ( $P > 0.73$ ), QT<sub>100</sub> ( $P > 0.33$ ), or QT<sub>10:100</sub> ( $P > 0.34$ ; Fig. 3) at exercise termination between the first and second time-trials.

During recovery, all indices remained significantly reduced compared with baseline up to 15 minutes following the second 1-min and 10-min time-trials ( $P < 0.05$ , Fig. 4, Table 1). In both groups, the rate of QT<sub>single</sub> recovery during the first and second recovery periods was lower after the first 10-min time-trial compared with the first 1-min time-trials and after the second compared with the first 1-min time-trial ( $P < 0.05$ , SDC 2). The rate of VA recovery during the first recovery period was higher after the 10-min compared with 1-min time-trials ( $P < 0.05$ ). In moderately trained cyclists, the rates of maximal voluntary contraction peak force and QT<sub>10:100</sub> recovery were lower after the second compared with the first time-trials, respectively (all  $P < 0.05$ ).

During the first recovery period and regardless of exercise duration, the rates of QT<sub>single</sub> and QT<sub>10:100</sub> recovery were lower in professional compared with moderately trained cyclists. This between-group difference persisted for QT<sub>10:100</sub> during the second recovery period after the first 1-min time-trial (all  $P < 0.05$ ). The rate of recovery of MVC peak force was also lower in professional compared with moderately trained cyclists after the first 1-min time-trial ( $P < 0.05$ ).

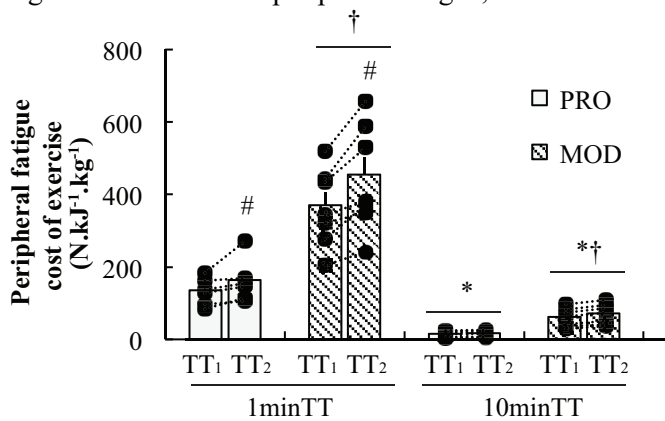
*Systemic response to exercise*

Mean  $\dot{V}O_2$  ( $P < 0.001$ ,  $d > 3.2$ ),  $\dot{V}CO_2$  ( $P < 0.001$ ,  $d > 3.2$ ), and breathing frequency ( $P < 0.01$ ,  $d > 1.87$ ) were significantly higher in professional compared with moderately trained cyclists in every time-trial, whereas minute ventilation ( $\dot{V}_E$ ) was higher in the 10-min time-trials only ( $P < 0.05$ ,  $d > 1.8$ ).  $\dot{V}_E/\dot{V}CO_2$  ( $P < 0.05$ ,  $d > 1.3$ ) and heart rate ( $P < 0.05$ ,  $d > 1.2$ ) were significantly lower in professional compared with moderately trained cyclists in the 1-min time-trials (SDC 3). No difference in capillary blood lactate concentration ( $P > 0.26$ ,  $d < 0.53$ ; SDC 3) was observed between groups in the 1-min time-trials or 10-min time-trials. Similarly, no effect of group, cycling time-trial duration or repetition on the rate of perceived exertion was found ( $P > 0.49$ ,  $d < 0.36$ ; SDC 4).

**DISCUSSION**

We sought to determine the recovery from neuromuscular fatigue following repeated cycling time-trials of different durations/intensities in professional and moderately trained cyclists. The improved time-trial

performance in professional cyclists was associated with a greater resistance to peripheral fatigue, as evidenced by



**Figure 5. Peripheral fatigue cost of exercise.**

Data are presented as mean group (bars) and individual (dots) data. The peripheral fatigue cost of exercise was calculated as follows:  $QT_{\text{single}}/W$ , where  $QT_{\text{single}}$  represents the change in  $QT_{\text{single}}$  from baseline to 15 s post-exercise (in N) and  $W$  the total work done during the corresponding exercise (in  $\text{kJ}\cdot\text{kg}^{-1}$ ). PRO, professional cyclists; MOD, moderately trained cyclists; \*  $P < 0.05$  between 1-min and 10-min time-trial; #  $P < 0.05$  between the first and second time-trial of the corresponding duration; †  $P < 0.05$  between PRO and MOD. The horizontal line encompasses time-points for which significant statistical differences between the first and second time trial or between groups were found.

a lower reduction in the potentiated twitch force in professional compared to moderately trained cyclists. Therefore, the peripheral fatigue cost of exercise was drastically reduced in these athletes. The time-course of recovery of the potentiated twitch force was slower in professional cyclists during its early phase, suggesting that peripheral fatigue recovery was driven by different mechanisms in the two groups. In addition, in both groups, exercise intensity, duration and the repetition of a given time-trial modulated the level of neuromuscular fatigue at the end of exercise and/or the time-course of recovery from neuromuscular fatigue.

### Neuromuscular fatigue at exercise termination

Our results showed that, following whole-body exercise, the ability to voluntarily produce a maximal force was less impaired in professional compared with moderately trained cyclists, regardless of the exercise intensity/duration and the level of pre-existing neuromuscular fatigue (Fig. 3). This can be mainly attributed to a preserved contractile function, as evidenced by the lower reductions in quadriceps twitch force indices in professional cyclists (Fig. 3). The lower peripheral fatigue in professional compared with moderately trained cyclists, together with the higher power output during every time-trials, resulted in a significantly lower peripheral fatigue cost of exercise in these athletes (Fig. 5). Given that  $QT_{10:100}$  was less reduced in the professional compared with moderately

trained cyclists and that quadriceps M-wave peak-to-peak amplitude remained unchanged in both groups, the lower peripheral fatigue in the professional cyclists was likely the consequence of preserved excitation-contraction coupling function (31). Our findings also showed that the contraction time and maximal rate of force development from  $QT_{\text{single}}$ , the degree of  $QT_{\text{potentiation}}$  and the peripheral fatigue cost of exercise were significantly lower in the professional compared with moderately trained cyclists. Together these results suggest a greater proportion of slow-twitch fatigue-resistant muscle fibers in professional cyclists' quadriceps (32,33), which would explain, at least in part, their preserved excitation-contraction coupling. Chronic endurance training is also associated with long-term adaptations within myocytes, which may have limited the accumulation of the intramuscular metabolites associated with peripheral fatigue (e.g.,  $P_i$ ,  $H^+$ , etc.) or reduced the myofibrillar sensitivity to a given concentration of these metabolites during exercise (16,34,35). These adaptations include, but are not limited to, greater reliance on oxidative metabolism (16), greater oxidative enzymatic activity (34), and/or reduced exercise-induced fragmentation of ryanodine receptors (35). Finally, in contrast to professional cyclists, MVC was further depressed at the end of the second compared to the first 1-min time-trial, in moderately trained cyclists. This further reduction in MVC, which occurred in the absence of any further reduction in  $QT_{\text{single}}$ , can be explained by an increase in central fatigue from the first to the second time-trial, as evidenced by the further and marked reduction in VA following the second 1-min time-trial (Fig. 3).

Consistent with previous findings (22,36), our results showed that, in both professional and moderately trained cyclists, the magnitude of neuromuscular fatigue is intensity-dependent. The reductions in potentiated twitch indices and VA were, respectively, 10-56% larger and 7-10% lower following the 1-min compared with 10-min time-trials. Our finding of no difference in  $QT_{10:100}$  between the two exercise durations suggests that the greater reduction in potentiated twitch force (i.e. peripheral fatigue) following the 1-min time-trials was not the result of a larger excitation-contraction coupling failure. Conversely, our results showing that normalized EMG amplitude increased by 37% to 56% during 1-min compared with 10-min time-trials and that this increase was significantly correlated to the further reduction in  $QT_{\text{single}}$  suggest that the higher level of peripheral fatigue was associated with the recruitment and fatigue of additional motor units (37). It is also consistent with previous muscle biopsy findings showing that the increase in muscle activation during a cycling time-trial led to a rise in the intramuscular metabolites associated with neuromuscular fatigue (e.g.,  $P_i$ ,  $H^+$ , ADP) and a greater degree of peripheral fatigue (9). In contrast, Schäfer et al. (23) found no difference between exercise-induced reduction in the potentiated twitch force when participants performed severe intensity exercise lasting 3-

Table 1. Effects of 1- and 10-min cycling time trials on neuromuscular fatigue indices

Indices	Group	1 <sub>min</sub> TT			10 <sub>min</sub> TT			TT <sub>2</sub>		
		Pre	15s	4min	Pre	15s	4min	15s	4min	4min
MVC (N)	PRO	577 ± 79	506 ± 52*	509 ± 75*	492 ± 49*	481 ± 88*#	555 ± 72	457 ± 62*	487 ± 51*	462 ± 56*#
	MOD	639 ± 76	523 ± 78*†	563 ± 103†	452 ± 54*#	457 ± 141*#	635 ± 118	437 ± 67*	496 ± 82*	456 ± 78*#
VA (%)	PRO	94 ± 5	89 ± 5†	90 ± 7*	87 ± 5*†	84 ± 10*#	93 ± 3	82 ± 6*	87 ± 6*	83 ± 9*#
	MOD	92 ± 4	89 ± 5†	86 ± 6*	84 ± 6*#†	80 ± 9*#	90 ± 3	80 ± 6*	82 ± 4*	80 ± 4*#
QT <sub>single</sub> (N)	PRO	166 ± 22	95 ± 24*†	116 ± 25*	91 ± 26*†	97 ± 24*#†	169 ± 21	120 ± 36*	121 ± 26*	119 ± 26*
	MOD	230 ± 40‡	84 ± 31*†	158 ± 45*† ‡	80 ± 28*†	105 ± 44*#†	234 ± 37‡	112 ± 34*	134 ± 47*	122 ± 37*#
QT <sub>10</sub> (N)	PRO	249 ± 29	148 ± 34*	163 ± 43*	148 ± 36*	147 ± 35*#	258 ± 33	161 ± 52*	166 ± 39*	158 ± 35*
	MOD	316 ± 52‡	106 ± 28*† ‡	199 ± 43*†	100 ± 25*† ‡	129 ± 52*#	322 ± 64‡	126 ± 39*	160 ± 59*	140 ± 55*#
QT <sub>100</sub> (N)	PRO	242 ± 20	186 ± 27*†	215 ± 33*	184 ± 26*†	202 ± 31*#	243 ± 21	200 ± 38*	211 ± 33*	206 ± 34*
	MOD	297 ± 47‡	178 ± 29*†	252 ± 44*	173 ± 34*†	197 ± 49*#†	303 ± 56‡	221 ± 35*	238 ± 49*	227 ± 47*#
QT <sub>10:100</sub>	PRO	1.0 ± 0.1	0.8 ± 0.1*	0.8 ± 0.1*	0.8 ± 0.1*	0.7 ± 0.1*	1.1 ± 0.1	0.8 ± 0.1*	0.8 ± 0.1*	0.8 ± 0.1*
	MOD	1.1 ± 0.1	0.6 ± 0.1*†	0.8 ± 0.1*#†	0.6 ± 0.1*†	0.6 ± 0.1*#	1.1 ± 0.1	0.6 ± 0.1*†	0.7 ± 0.2*	0.6 ± 0.2*†
CT (ms)	PRO	68 ± 6	67 ± 7†	66 ± 1†	65 ± 6†	64 ± 3†	68 ± 6	59 ± 5*	60 ± 3*	59 ± 3*
	MOD	59 ± 3‡	63 ± 5†	59 ± 4‡	61 ± 4†	61 ± 4† ‡	60 ± 6‡	58 ± 6*	58 ± 4*	56 ± 5*
MRFD (N.s <sup>-1</sup> )	PRO	2.5 ± 0.5	1.4 ± 0.3*†	1.8 ± 0.4*†	1.5 ± 0.4*†	1.6 ± 0.4*†	2.5 ± 0.4	2.0 ± 0.5*	2.0 ± 0.4*	2.0 ± 0.4*
	MOD	3.9 ± 0.7‡	1.4 ± 0.6*†	2.7 ± 0.9*† ‡	1.3 ± 0.4*†	1.7 ± 0.7*#†	3.9 ± 0.6‡	2.0 ± 0.6*	2.3 ± 0.8*	2.3 ± 0.8*
HRT (ms)	PRO	81 ± 14	80 ± 13†	63 ± 9*†	77 ± 14†	63 ± 5*† ‡	88 ± 13	67 ± 6*	54 ± 4*	46 ± 4*#
	MOD	62 ± 11‡	68 ± 19†	57 ± 8†	68 ± 10†	53 ± 4*† ‡	68 ± 16‡	48 ± 11*†	42 ± 6*†	37 ± 6*†
VL M <sub>MAX</sub> (mV)PRO		20 ± 4	22 ± 3	22 ± 4	23 ± 3	22 ± 4	19 ± 3	19 ± 3	19 ± 3	19 ± 2
	MOD	17 ± 5	17 ± 5	18 ± 5	18 ± 5	18 ± 5	16 ± 4	16 ± 4	16 ± 4	15 ± 4
VM M <sub>MAX</sub> (mV)PRO		24 ± 5	23 ± 4	25 ± 6	24 ± 5	26 ± 6	23 ± 5	23 ± 4	24 ± 4	23 ± 4
	MOD	23 ± 6	22 ± 6	24 ± 5	22 ± 6	23 ± 5	20 ± 5	20 ± 5	21 ± 5	19 ± 5
RF M <sub>MAX</sub> (mV)PRO		5.4 ± 1.1	5.3 ± 1.1	5.7 ± 1.0	5.5 ± 1.1	5.8 ± 0.6	5.5 ± 1.2	5.4 ± 0.9	5.7 ± 0.8	5.2 ± 0.3
	MOD	4.1 ± 0.8	4.1 ± 1.0	4.6 ± 1.3	4.4 ± 1.1	4.3 ± 0.9	4.7 ± 1.8	4.9 ± 1.8	5.1 ± 1.7	4.7 ± 1.3

Data are expressed as mean ± SD in absolute units. CT, MRFD and HRT were calculated from QT<sub>single</sub>, PRO, professional cyclists; MOD, moderately trained cyclists; MVC, maximal voluntary contraction; VA, voluntary activation of the quadriceps; QT<sub>single</sub>, potentiated quadriceps twitch evoked with supramaximal electrical single stimulation; QT<sub>10</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 10-Hz paired stimulation; QT<sub>100</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 100-Hz paired stimulation; QT<sub>10:100</sub>, low-frequency fatigue ratio (QT<sub>10</sub> / QT<sub>100</sub>); CT, contraction time; MRFD, maximal rate of force development; HRT, half relaxation time; VL M<sub>MAX</sub>, maximal amplitude of the compound muscle action potential for the *vastus lateralis*; VM M<sub>MAX</sub>, maximal amplitude of the compound muscle action potential for the *vastus medialis*; RF M<sub>MAX</sub>, maximal amplitude of the compound muscle action potential for the *rectus femoris*; 10<sub>min</sub>TT, 10-min time-trial; 1<sub>min</sub>TT, 1-min time-trial; I<sub>min</sub>TT, 1-min time-trial; I<sub>min</sub>TT<sub>1</sub> vs. 10<sub>min</sub>TT<sub>1</sub> or 1<sub>min</sub>TT<sub>2</sub> vs. 10<sub>min</sub>TT<sub>2</sub>; † P < 0.05 vs. Pre; # P < 0.05 vs. I<sub>min</sub>TT<sub>1</sub> vs. 10<sub>min</sub>TT<sub>1</sub> or 1<sub>min</sub>TT<sub>2</sub> vs. 10<sub>min</sub>TT<sub>2</sub>; ‡ P < 0.05 vs. PRO

or 12-min, until complete depletion of the total amount of work performed above the critical power (23). It has also been shown that the end-exercise change in metabolic variables associated with peripheral fatigue (e.g.  $H^+$ ,  $K^+$ ) were similar when ~2 to ~14-min cycling exercises were performed to exhaustion (10). These biopsy data indirectly suggest that the degree of peripheral fatigue – which is mainly determined by the concentration level of metabolic by-products associated with muscle fatigue – might be similar following severe intensity exercise of various duration. The discrepancy on the effect of exercise intensity / duration in the severe intensity domain on exercise-induced neuromuscular fatigue between our and the two above-mentioned findings might be explained by 1) the delay to the first neuromuscular measurement after exercise termination that was shortened to 15-s in our study compared to 1-min in Schäfer et al.'s study (vs. 1-min in Schäfer et al., 23). This short delay allowed us to capture a larger component of the rapid fatigue recovery processes which take place during the first 30-s following exercise (see Fig. 4 and 13, 38-40), therefore limiting the risk of an underestimation of exercise-induced neuromuscular fatigue. 2) The increase in EMG amplitude in response to the higher force demand with the increase in power output in our study (vs. no change in Black et al., 10), which is likely indicative of the recruitment and subsequent fatigue of a greater portion of the motoneuron pool. A limitation to these explanations is an acknowledgement that estimate of muscle activation from surface EMG amplitude is, despite its extensive use in the literature, subject to a number of valid critiques. This includes, but is not limited to, motion artifacts, motor-units amplitude cancellation, muscle crosstalk, and/or changes in conduction velocity (41-43).

In both professional and moderately trained cyclists, the reduction in potentiated twitch indices was similar at the end of the two 1- and 10-min time-trials, despite very different power outputs, EMG amplitudes and pre-existing degrees of peripheral fatigue. These results are consistent with the concept that muscle activation and power output are regulated during whole-body exercise to limit the development of peripheral fatigue – or more likely the concentration of the intramuscular metabolites associated with neuromuscular fatigue – at an individual and constant level (8,9,12,14,15). Specifically, the stimulation of group III-IV afferent nerve endings by the intramuscular accumulation of metabolic by-products (44-48) and mechanical stimuli during exercise (49,50), inhibits muscle activation to prevent an abnormal and potentially harmful impairment of the contracting muscle as a result of a severe disturbance of the intramuscular metabolic milieu (8,9,51,52). Our findings now provide evidence that this 'threshold' of peripheral fatigue is modulated by exercise intensity and duration and occurs at a lower level of peripheral fatigue in professional compared with moderately trained cyclists (Fig. 3).

### Recovery from neuromuscular fatigue

In the present study, we reduced the delay between the end of a whole-body exercise and the neuromuscular function tests to exactly 15 s and were thus able to accurately capture the time-course and determine the etiology of recovery from exercise-induced neuromuscular fatigue in professional and moderately trained cyclists. Our original findings showed that, although voluntary or evoked force did not differ between the professional and moderately trained cyclists 15-min following the second time-trials (Fig. 4), the recoveries from neuromuscular fatigue had different time-courses and involved different mechanisms in the two groups. The differences were mainly found within the first two minutes following the end of exercise (i.e., the first recovery period), with professional cyclists showing a slower rate of peripheral fatigue recovery than moderately trained cyclists (Fig. 4, SDC 2). The early phase of twitch force recovery following severe-intensity exercise is in great part driven by the washout of metabolites associated with phosphocreatine hydrolysis and ATP depletion (i.e., inorganic phosphate and adenosine diphosphate) (53-56). We thus interpret our findings as indirect evidence that the accumulation of these metabolites during exercise and the consecutive need for washout following exercise were much lower in professional cyclists. This hypothesis is supported by our findings suggesting a greater proportion of slow-twitch and mainly oxidative muscle fibers within professional cyclists' quadriceps (see previously), as well as our metabolic and cardioventilatory data (SDC 3) indicating that the contribution of the oxidative pathways to ATP resynthesis was higher in professional compared with moderately trained cyclists. Moreover, the absence of  $QT_{10:100}$  recovery in professional cyclists after all cycling time-trials indicates that peripheral fatigue recovery was influenced by the long-lasting effects of the processes associated with prolonged low-frequency force depression (57,58). Prolonged low-frequency force depression following cycling exercise in the severe intensity domain might be explained, at least in part, by decreased myofibrillar sensitivity to  $Ca^{2+}$  and/or decreased amount of  $Ca^{2+}$  released from the sarcoplasmic reticulum in response to the exercise-induced accumulation of reactive oxygen species (57-59).

In both groups, the  $QT_{single}$  recovery in the first two periods of recovery (i.e., from 15-s to 4-min) was significantly faster following the 1-min than the 10-min time-trials. The enhanced muscle activation and power output during the 1-min time-trials (Fig. 2) were likely associated with a greater contribution of fast-twitch, fatigue-sensitive muscle fibers to force production. During intense exercise, these fibers consume high amounts of ATP and creatine phosphate and consequently accumulate inorganic phosphate and adenosine diphosphate (60), two metabolites associated with peripheral fatigue (3). We thus speculate that the

faster time-course of recovery from peripheral fatigue during the 1-min compared with 10-min time-trials was linked to the rapid time-course of phosphagen resynthesis and metabolite clearance within the locomotor muscles (53-56).

Recovery from central fatigue showed a different pattern and time-course after the 1-min compared with 10-min time-trials. While VA returned to baseline after the 10-min time-trials, it progressively decreased during the first recovery period (i.e., up to 2-min) following the 1-min time-trials. This remarkable result was observed in every participant and after both the first and second 1-min time-trials. It is thus unlikely a measurement artifact. While only very few studies have been designed to capture the time-course of central fatigue recovery during its early phase following whole-body exercise, a reduction in VA during recovery was found following 10- to 30-s all-out sprints (13,61). This reduction in VA over time can be prevented when the projections of the group III-IV muscle afferents are attenuated using pharmacological blockade (62). Conversely, VA recovery can be blunted when the tonic input from group III-IV muscle afferents is maintained at its exercise level by trapping the metabolites released during exercise within the myocytes using arterial occlusion (56,63). We thus speculate that group III-IV muscle afferent feedback rose during the early phase of recovery following the 1-min time-trials in response to the accumulation of metabolites like  $H^+$  (54,64) and contributed to accentuating central fatigue during recovery. Further studies are nevertheless needed to verify this hypothesis.

Last, repeating the cycling time-trials significantly slowed the time-course of recovery from peripheral fatigue, while the effect on central fatigue was more limited. Consistent with our findings are results showing that peripheral fatigue recovery was slowed following repeated all-out 10-s cycling sprints (13). The slower  $QT_{10:100}$  recovery after the second compared with the first 1-min and 10-min time-trials suggests that the magnitude of the prolonged low-frequency force depression might have increased following the second time-trial. This likely contributed to the increase in the peripheral cost of exercise (Fig. 5) and delayed recovery from peripheral fatigue (57,58).

## CONCLUSION

Our original findings showed that the improved performances of professional compared with moderately

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trained cyclists were associated with greater resistance to peripheral fatigue during repeated cycling time-trials of various intensities and durations. Remarkably, professional cyclists showed a slower rate of peripheral fatigue recovery during its early phase, suggesting that the combined metabolites accumulation and neuromuscular fatigue during exercise and the consecutive need for metabolite clearance following exercise were much lower in these athletes. Rather, the time-course of recovery from peripheral fatigue in professional cyclists was dependent on the long-lasting effects of prolonged low-frequency force depression. In both groups, the increase in exercise intensity and the repetition of a given time-trial increased the peripheral cost of exercise and modulated the time-course of neuromuscular fatigue recovery. The findings of this study enhance our understanding of the etiology of neuromuscular fatigue and the recovery in elite and amateur endurance athletes. They therefore provide important insights about the neuromuscular limits of human performance and have applications in training load management in the exercising human.

## ADDITIONAL INFORMATION

The results of the study are presented clearly, honestly, and without fabrication, falsification, or inappropriate data manipulation, and statement that results of the present study do not constitute endorsement by ACSM.

## COMPETING INTEREST

The authors have no conflicting interests

## AUTHOR CONTRIBUTIONS

G.P.D and G.M.B. conceived the study, interpreted the data and prepared the manuscript. G.P.D. analyzed the data. All authors executed the study and edited, revised and approved the final version of the manuscript. All authors agree to be accountable for all aspects of the work, ensuring that questions related to the accuracy or integrity of any part are appropriately investigated and resolved. All persons designated as authors qualify for authorship, and all those who qualify for authorship are listed.

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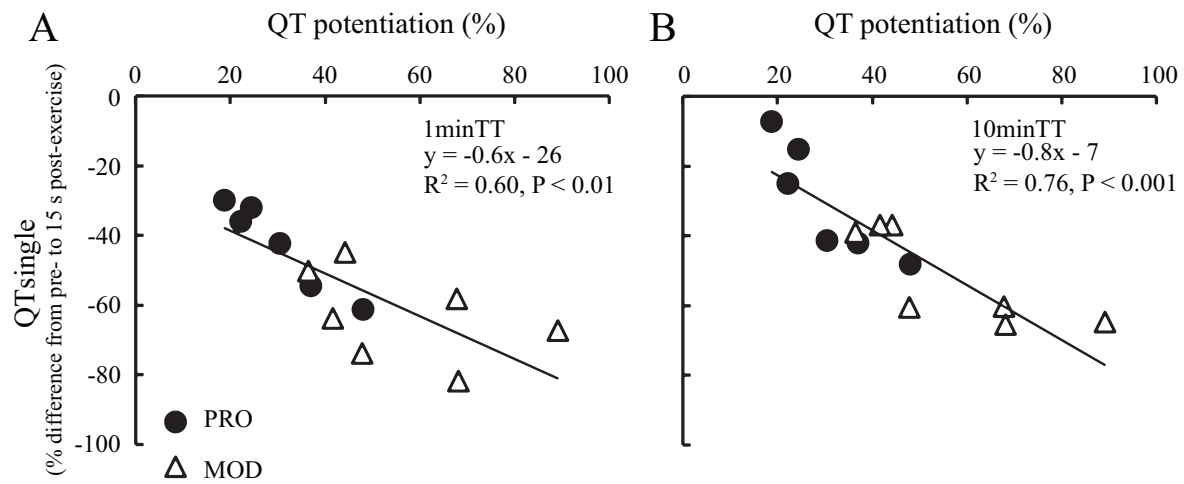
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**SDC 1. Relationship between quadriceps fatigue and post-activation potentiation.**

Pre- to 15-s post-exercise difference in potentiated quadriceps twitch evoked with supramaximal electrical single stimulation ( $QT_{single}$ ) and quadriceps post-activation potentiation (QT potentiation) from the first ten minute (A) and the first one minute (B) cycling time-trials are presented. PRO, professional cyclists; MOD, moderately-trained cyclists.

**SDC 2. Rates of recovery from fatigue in professional and moderately trained cyclists**

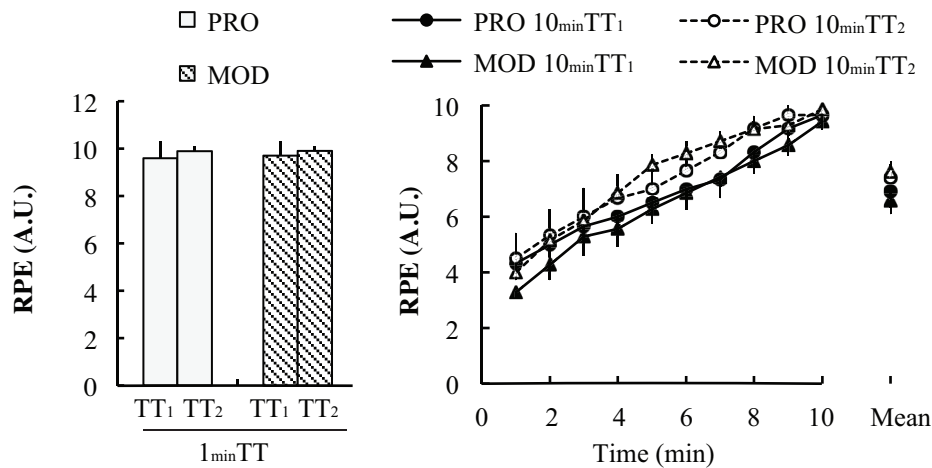
Indice (%/min)	Group	1minTT						10minTT					
		TT1		TT2			TT1		TT2				
		15s-2min	2-4min	15s-2min	2-4min	4-15min	15s-2min	2-4min	15s-2min	2-4min	4-15min		
MVC	PRO	-17 ± 18	15 ± 16 *	-10 ± 20	6 ± 8 *	2 ± 2	7 ± 17 †	7 ± 20	0 ± 18	6 ± 10	1 ± 3		
	MOD	11 ± 19 ‡	14 ± 9	1 ± 18 #	4 ± 8 #	4 ± 2	19 ± 23	3 ± 4	1 ± 20 #	0 ± 8 #	5 ± 3 ‡		
VA	PRO	-5 ± 10	5 ± 5	-7 ± 11	5 ± 12	1 ± 2	10 ± 10 †	4 ± 7	8 ± 15 †	2 ± 3	2 ± 3		
	MOD	-13 ± 9	2 ± 11	-15 ± 11	4 ± 9 *	2 ± 1	6 ± 13 †	1 ± 6	4 ± 12 †	-1 ± 3	1 ± 1		
QT <sub>single</sub>	PRO	9 ± 7	5 ± 6	3 ± 5 #	1 ± 3 #	1 ± 1	-4 ± 19 †	0 ± 5 †	-1 ± 16	-2 ± 2	-1 ± 1		
	MOD	19 ± 5 ‡	9 ± 4 *	7 ± 6 #	3 ± 2 #	2 ± 1	9 ± 9 †	2 ± 1 *†	6 ± 7	0 ± 3	1 ± 2 ‡		
QT <sub>10</sub>	PRO	6 ± 7	1 ± 3	1 ± 5 #	-2 ± 3	0 ± 1	4 ± 6	-2 ± 5	0 ± 8 #	-1 ± 5	-1 ± 1		
	MOD	17 ± 6 ‡	7 ± 4 *‡	7 ± 6 #‡	2 ± 3 *#‡	1 ± 0 ‡	10 ± 6 †‡	1 ± 2 *†	4 ± 5 #	0 ± 2	1 ± 2		
QT <sub>100</sub>	PRO	14 ± 17	5 ± 10	10 ± 17	2 ± 7	-1 ± 2	11 ± 29	10 ± 23	6 ± 20	-1 ± 6	-1 ± 5		
	MOD	25 ± 7	11 ± 7 *	9 ± 10 #	5 ± 2	3 ± 1	9 ± 17 †	4 ± 7	4 ± 16	3 ± 12	2 ± 1		
QT <sub>10:100</sub>	PRO	-1 ± 12	-2 ± 5	-6 ± 8	-4 ± 4	0 ± 2	3 ± 13	-3 ± 7	-4 ± 9	-1 ± 7	-1 ± 2		
	MOD	18 ± 10 ‡	4 ± 4 *‡	9 ± 5 #‡	0 ± 4 *	0 ± 1	12 ± 5 ‡	-1 ± 3 *	5 ± 5 #‡	-1 ± 5	1 ± 3		

Data are expressed as mean ± SD. A negative value indicates fatigue further developed during the given recovery period. PRO, professional cyclists; MOD, moderately-trained cyclists; MVC, maximal voluntary contraction; VA, voluntary activation of the quadriceps; QT<sub>single</sub>, potentiated quadriceps twitch evoked with supramaximal electrical single stimulation; QT<sub>10</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 10-Hz -paired stimulation; QT<sub>100</sub>, potentiated quadriceps twitch evoked with supramaximal electrical 100-Hz paired stimulation; QT<sub>10:100</sub>, low-frequency fatigue ratio (QT<sub>10</sub> / QT<sub>100</sub>); 10<sub>min</sub>TT, 10-minute time-trial; 1<sub>min</sub>TT, 1-minute time-trial; TT<sub>1</sub>, first time-trial, TT<sub>2</sub>, second time-trial; \*, *P* < 0.05 vs. the preceding time period # *P* < 0.05 vs. first time-trial; † *P* < 0.05 vs. 10<sub>min</sub>TT<sub>1</sub> vs. 1<sub>min</sub>TT<sub>1</sub> or 10<sub>min</sub>TT<sub>2</sub> vs. 1<sub>min</sub>TT<sub>2</sub>; ‡ *P* < 0.05 vs. PRO.

**SDC 3. Metabolic and cardioventilatory responses to 1- and 10-minute cycling time trials.**

Indices	1 <sub>min</sub> TT				10 <sub>min</sub> TT			
	TT <sub>1</sub>		TT <sub>2</sub>		TT <sub>1</sub>		TT <sub>2</sub>	
	PRO	MOD	PRO	MOD	PRO	MOD	PRO	MOD
$\dot{V}O_2$ (ml.min <sup>-1</sup> .kg <sup>-1</sup> )	52 ± 7	37 ± 6‡	58 ± 5#	45 ± 7#‡	69 ± 5†	50 ± 8†‡	65 ± 6#†	47 ± 7#†‡
$\dot{V}CO_2$ (ml.min <sup>-1</sup> .kg <sup>-1</sup> )	57 ± 4	41 ± 9‡	51 ± 4#	40 ± 6‡	66 ± 5†	50 ± 8†‡	54 ± 6#†	40 ± 5#‡
$\dot{V}CO_2 \cdot \dot{V}O_2^{-1}$	1.1 ± 0.1	1.1 ± 0.1	1.0 ± 0.1#	0.9 ± .02#	1.0 ± .03†	1.0 ± 0.2†‡	0.8 ± 0.1#†	0.9 ± .02#
$\dot{V}_E$ (l.min <sup>-1</sup> )	130 ± 21	109 ± 42	153 ± 21#	142 ± 31#	146 ± 15†	114 ± 30†‡	141 ± 20†	114 ± 25†‡
$f_B$ (breath.min <sup>-1</sup> )	53 ± 10	40 ± 10	58 ± 10#	54 ± 7#	49 ± 5†	38 ± 7‡	53 ± 4#†	45 ± 8#†
$V_T$ (l)	2.4 ± 0.2	2.8 ± 1.0	2.6 ± 0.2#	2.6 ± 0.4	3.0 ± 0.3†	3.0 ± 0.4	2.7 ± 0.4#†	2.5 ± 0.4#
HR (beats.min <sup>-1</sup> )	155 ± 9	168 ± 14	158 ± 9#	170 ± 12	172 ± 8†	175 ± 6†	178 ± 6†#	181 ± 6#†
[La] <sub>b</sub> (mmol.l <sup>-1</sup> )	13.6 ± 3	14.4 ± 2.4	15.3 ± 2.7#	16.3 ± 1.9#	12.4 ± 2.2	13 ± 2.2	11 ± 3.4	12.6 ± 2.8

Data are expressed as mean ± SD.  $\dot{V}O_2^{-1}$ , oxygen uptake;  $\dot{V}CO_2^{-1}$ , carbon dioxide output;  $\dot{V}_E$ , minute ventilation;  $f_B$ , breath frequency;  $V_T$ , tidal volume; HR, heart rate; [La]<sub>b</sub>, capillary blood lactates; 10<sub>min</sub>TT, 10-minute time-trial; 1<sub>min</sub>TT, 1-minute time-trial; TT<sub>1</sub>, first time-trial, TT<sub>2</sub>, second time-trial; PRO, professional cyclists; MOD, moderately-trained cyclists. #  $P < 0.05$  vs. first time-trial; †  $P < 0.05$  10<sub>min</sub>TT<sub>1</sub> vs. 1<sub>min</sub>TT<sub>1</sub> or 10<sub>min</sub>TT<sub>2</sub> vs. 1<sub>min</sub>TT<sub>2</sub>; ‡  $P < 0.05$  vs. PRO.



**SDC 4. Rate of perceived exertion (RPE) estimated at the end of the 1-min time-trials and during the 10-min time-trials.**

Data are presented as mean  $\pm$  SE. RPE was measured right before exercise cessation the end in 1-minute time-trials (1minTT) and 10-minute time-trials (10minTT), as well as every minute during exercise in 10minTTs. PRO, professional cyclists; MOD, moderately trained cyclists; TT<sub>1</sub>; first time-trial, TT<sub>2</sub>, second time-trial. No effect of group (PRO vs. MOD), time-trial duration (RPE at exercise termination in 1minTTs vs. 10minTTs) or repetition (TT<sub>1</sub> vs. TT<sub>2</sub>) were found.