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Spectral properties of muscle activation during incremental cycling test

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Abstract

It is uncertain whether fatigue and workload would affect muscle recruitment during cycling. To infer on changes in priority for recruitment of motor units, we assessed the combined effects of fatigue and workload during an incremental cycling test to exhaustion on frequency components of lower limbs muscle activation. Competitive cyclists performed an incremental maximal cycling test while muscle activations were recorded from the right and left vastus lateralis, biceps femoris and gastrocnemius medialis. Muscle activation signals were assessed using frequency band analyses and decomposed into overall, high and low frequency bands. Combined effects from workload/fatigue were assessed using t tests and Cohen's effect sizes (ES). There were increases in the overall muscle activation due to increased workload/fatigue for biceps femoris (40% vs. 90%, p<0.01 and ES = 1.85) and vastus lateralis (40% vs. 70%, p=0.01 and ES = 0.96, and 40% vs. 90%, p<0.01 and ES = 2.03, and 70% vs. 90%, p<0.01 and ES = 0.96), but not for gastrocnemius medialis. There was also greater contribution from low frequency component for biceps femoris (40% vs. 90%, p = 0.01 and ES = 1.12). Similar workload/fatigue effects have been observed between lower limbs. In conclusion, incremental cycling test lead to an increase in activation of main knee joint flexors and extensors but not in plantar flexors during cycling. Biceps femoris changes its recruitment profile due to increases in low frequency content.

Keywords: testing, fatigue, motor control, electromyography, frequency band analysis, cyclists.

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Introduction

Muscle activation during cycling increases with sub maximal fatigue or workload increments (Dorel et al. 2009; Priego et al. 2014), which has been described using time series analyses signals recorded using surface electromyography (EMG) (Hug and Dorel 2009). Muscle activation is affected by changes in firing rates of each motor unit and by the number of motor units recruited by the central nervous system (Wakeling and Horn 2009). Decomposition of muscle activation signals provide insight into the main sources of changes in muscle recruitment during fatigue that are not detectable using time series analysis (Wakeling and Horn 2009). Indeed, during cycling, fatigue changes spectral properties of EMG signals and potentially priority in motor unity recruitment (von Tscharner 2002; Diefenthaeler et al. 2012a). However, the time course of this adaptation during exercise is still under debate. During mild fatigue cycling exercise, low frequency components of gastrocnemius medialis and the vastus lateralis activation increases (Von Tscharner 2009), while cycling to exhaustion reduces high frequency components of vastus lateralis and biceps femoris activation, which suggests that priority in motor unity recruitment may be affected by fatigue (Diefenthaeler et al. 2012a).

Knee and hip extensor muscles play an important role for force production acting as "muscle drivers" during pedalling (Fregly and Zajac 1996). However, other muscles, such as plantar flexors, act as energy transfers from the limbs to the cranks and generate tangential force on the cranks (Fregly and Zajac 1996; Wakeling and Horn 2009). Although there is evidence that greater workloads linearly increase activation of main muscle drivers (Hug et al. 2004), it is unclear whether increases in muscle activation emerge from larger and/or smaller motor units. In response to increase in exercise workload, larger motor units should be recruited later than smaller motor units (Henneman 1957). In exercise fitness assessment, incremental load tests are used for performance assessment and physiological monitoring which provide information for training load prescription (Bentley et al. 2007). Given that some muscles present a threshold in activation profile during graded exercise tests (Hug et al. 2003b, 2006), it is likely that changes in priority in motor unity recruitment could be affected by the workload level. These could be linked to the potential off-set in priority of recruitment of large and small size motor units, which has not been assessed in previous studies.

The use of frequency bands to infer motor unit recruitment strategies and the fiber-type composition of a muscle is criticized by different authors stating that surface EMG power spectrum is influenced by other factors than the average conduction velocity of the action potentials (Amann et al. 2008; Farina 2008). These factors are mainly that amplitude cancellation and/or significant filtering effects can attenuate increases in motor unit activity measured by surface EMG and voluntary muscle contractions might alter the efficacy of corticospinal synapses on motor neurons (Amann et al. 2008; Farina 2008). However, some studies that used histochemical and contractile measures of fiber type supported an important association between the spectral properties of surface electromyography and motor unit recruitment strategies (Kupa et al. 1995; Wakeling and Syme 2002; Hodson-Tole and Wakeling 2007). This is the reason for why frequency band analysis may be useful for characterization of motor unit recruitment strategies and muscle fiber type (von Tscharner and Nigg 2008).

To investigate the combined effects of workload and fatigue on priority in motor unity recruitment, we compared frequency components of bilateral lower limb muscle activation during an incremented cycling exercise to exhaustion. During outdoor cycling and laboratorial tests, cyclists are submitted to a combination of increases in fatigue state and workload (Abbiss et al. 2006; Blake and Wakeling 2012) which potentially leads to changes in recruitment of motor units. We hypothesized that when cycling to exhaustion, cyclists present a different motor unit recruitment strategy in order to sustain force production. This mechanism should encompass greater contribution from high frequency components in performance-related muscles (i.e. vastus lateralis and biceps femoris) throughout the test along with maintenance in comparison with a force-transfer muscle (gastrocnemius medialis).

Materials and methods

Participants

Twelve male competitive cyclists participated in this study and signed an informed consent term in agreement with the local Committee of Ethics in Research with Humans and according to international standards (Harriss and Atkinson 2011). Cyclists were asked to avoid high-intensity or exhaustive exercises at least 24 h before the laboratory trials. The mean and standard deviation values for age, body mass, height, maximal oxygen uptake (VO_{2MAX}), peak power output, and power mass ratio of the participants were 27 ± 6.3 years, 77 ± 10.0 kg, 177 ± 8.7 cm, 58 ± 7.3 $ml \cdot kg^{-1} \cdot min^{-1}$, 360 ± 50 W, and 4.7 ± 0.8 $W \cdot kg^{-1}$, respectively. Three cyclists had the left limb preferred according to Waterloo inventory (Elias et al. 1998).

Table 1. Frequency bands selected from previous non-continuous wavelet model (von Tscharner 2002) used on the series of band-pass digital filtering of EMG signals of each muscle (Diefenthaeler et al. 2012a; Priego et al. 2014).

Band	1	2	3	4	5	6	7	8	9
High	48.45	75.75	110.00	149.00	193.45	244.45	300.80	363.80	431.65
Low	26.95	48.45	74.80	108.00	146.95	191.75	242.20	297.40	359.35

Testing

Cyclists underwent an incremental maximal cycling test to exhaustion using their own bicycles attached to a cycle trainer (Computrainer ProLab, RacerMate, Inc., Seattle, MA, USA). The incremental cycling test started at a workload level of 50 W with progressive increments of 25 W every minute until exhaustion (Lucía et al. 2002). Cyclists were asked to maintain their preferred pedaling cadence throughout the test. Exhaustion was defined as the moment when cyclists were no longer capable of maintaining the minimal cadence of 70 rpm. Maximal power output was defined as the workload of last stage completed. VO_{2MAX} was defined as the highest average oxygen uptake over a 30-s period.

Data acquisition

Gas exchange and EMG activation were monitored continuously throughout the entire incremental maximal test. Gas exchanges were captured using the breath-by-breath method by an open-circuit indirect gas exchange system (MGC CPX/D, Medical Graphics Corp., St Louis, MO, USA) to determine VO_{2MAX}. Muscle activation was monitored using surface EMG from the right and left vastus lateralis, biceps femoris and gastrocnemius medialis. Signals were amplified and recorded at 2000 Hz with 14-bit resolution (Miotool, Miotec Biomedical, Porto Alegre, RS, Brazil) for off-line signal processing. Pairs of Ag/AgCl electrodes (bipolar configuration) with a diameter of 22 mm (Kendall Meditrace, Chicopee, Canada) were positioned on the skin after careful shaving and cleaning of the area with an abrasive cleaner and alcohol swabs to reduce the skin impedance (De Luca 1997). A reference electrode placed over the skin of the acromion served as a neutral site. The electrodes were placed over the belly of the muscles, parallel with the muscle fiber orientation (Hermens et al. 2000) and taped to the skin using micropore tape (3M Company, St Paul, MN, USA) to minimize movement artifact.

Data analysis

The fully procedures of frequency bands analyses followed a discrete analysis of band-pass filtering (Diefenthaeler et al. 2012a; Priego et al. 2014). Each muscle's EMG signal was filtered using each of the nine combinations of high (193.45-300.80 Hz) and low (26.95-48.45 Hz) band stop (frequency bands) filters (Table 1). EMG signals of each of the nine frequency

Table 2. Comparison of EMG activation between the preferred (P) and the non-preferred limb (NP) for overall, high and low frequency of the different muscles (BF – biceps femoris, GM - gastrocnemius medialis, and VL - vastus lateralis) during the test. Positive values indicate greater activation for the preferred limb.

% test	OVERALL			HIGH			LOW		
	BFP vs. BFNP	GMP vs. GMNP	VLP vs. VLNP	BFP vs. BFNP	GMP vs. GMNP	VLP vs. VLNP	BFP vs. BFNP	GMP vs. GMNP	VLP vs. VLNP
40% **(%diff, ES and p)**	15 ±27%;	4 ±14%	8 ±30%	15 ±8%	-14 ±11%	12 ±6%	9 ±11%	-1 ±9%	-1 ±12%
	ES =0.77	ES =0.28	ES =0.42	ES =0.10	ES =0.50	ES =0.04	ES =0.17	ES =0.48	ES =0.24
	p = 0.07	p = 0.48	p = 0.12	p = 0.75	p = 0.23	p = 0.72	p = 0.62	p = 0.38	p = 0.01
70% **(%diff, ES and p)**	11 ±54%;	4 ±24%	9 ±68%	16 ±7%	-16 ±10%	12 ±5%	9 ±9%	2 ±8%	-4 ±10%
	ES =0.38	ES =0.19	ES =0.27	ES =0.11	ES =0.28	ES =0.10	ES =0.18	ES =0.47	ES =0.30
	p = 0.37	p = 0.65	p = 0.31	p = 0.74	p = 0.48	p = 0.34	p = 0.60	p = 0.36	p = 0.12
90% **(%diff, ES and p)**	13 ±62%;	1 ±31%	0 ±87%	8 ±6%	-13 ±10%	14 ±4%	3 ±7%	-3 ±10%	-1 ±11%
	ES =0.48	ES =0.03	ES =0.00	ES =0.29	ES =0.19	ES =0.38	ES =0.08	ES =0.12	ES =0.03
	p = 0.21	p = 0.95	p = 0.99	p = 0.51	p = 0.58	p = 0.13	p = 0.85	p = 0.12	p = 0.02

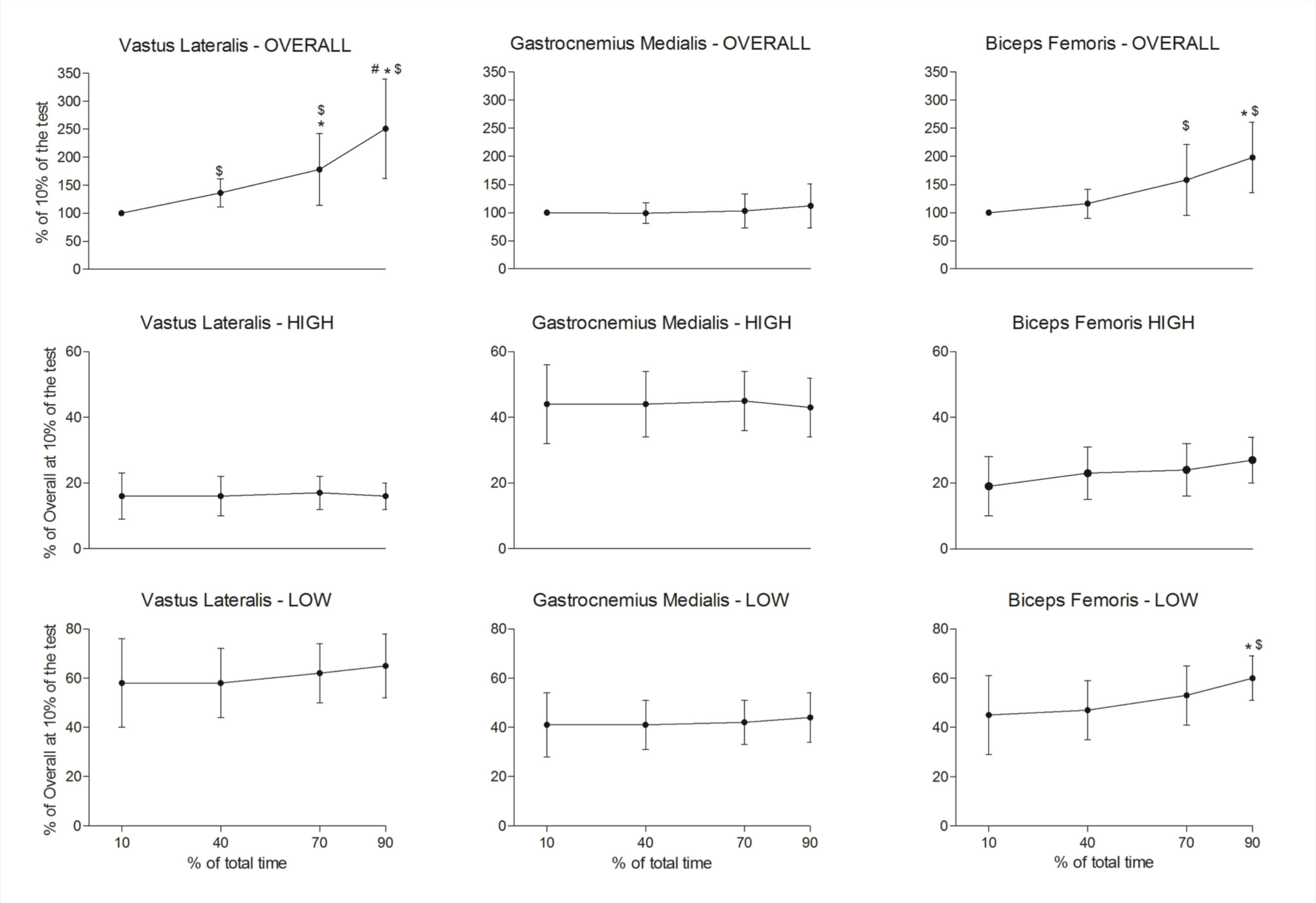

Figure 1. EMG activation normalized by the 10% of total time of the test. Overall, high and low frequency of the different muscles (BF – biceps femoris, GM - gastrocnemius medialis, and VL - vastus lateralis) in the preferred limb during the test. EMG was analyzed in four moments: 10% (averaged 10 to 20%), 40% (averaged 40 to 50%), 70% (averaged 70 to 80%) and 90% (averaged 90 to 100%). Differences are indicated when ES>0.80 and p<0.05 using different symbols depending on the differences in each time interval ($ - differences to 10% of total time, * - differences to 40% of total time, # - differences to 70% of total time).

bands that resulted from the filtering process were then converted into their root mean square values (RMS) computed using moving average windows of 40 ms (Neptune et al. 1997) to determine the intensity of muscle activation throughout the test.

Table 3. P values and effects sizes (ES) for differences between muscles (BF – biceps femoris, GM - gastrocnemius medialis, and VL - vastus lateralis) in the EMG frequency bands of the preferred limb during the test. EMG was analyzed in four instants: 10% (averaged 10 to 20%), 40% (averaged 0 to 50%), 70% (averaged 70 to 80%) and 90% (averaged 90 to 100%). Significant differences (p<0.05 and ES>0.8) are indicated using bold italics.

	VL	GM	BF	VL vs GM		VL vs BF		GM vs BF	
	Mean±SD(%)	Mean±SD(%)	Mean±SD(%)	p	ES	p	ES	p	ES
Overall									
40	136±25	99±18	116±26	.002	1.7	.06	0.8	.08	0.8
70	178±64	103±30	158±63	.003	1.6	.47	0.3	.01	1.2
90	251±89	112±39	198±63	<.001	2.2	.08	0.7	.001	1.7
High									
10	16±7	44±12	19±9	<.001	3.0	.24	0.5	<.001	2.3
40	16±6	44±10	23±8	<.001	3.4	.05	0.9	<.001	2.3
70	17±5	45±9	24±8	<.001	3.9	.007	1.1	<.001	2.4
90	16±4	43±9	27±7	<.001	4.2	<.001	2.0	<.001	2.1
Low									
10	58±18	41±13	45±16	.04	1.1	.02	0.8	.58	0.3
40	58±14	41±10	47±12	.005	1.4	.06	0.8	.18	0.6
70	62±12	42±9	53±12	<.001	2.0	.003	0.8	.002	1.1
90	65±13	44±10	60±9	.001	1.7	.12	0.4	<.001	1.6

The sum of the nine average frequency bands was calculated for the computation of power from the overall RMS of each muscle (i.e. activation of all frequency bands of the EMG signal). The fifth, sixth and seventh bands were averaged to compute the RMS from high frequency components of the signals, which would potentially represent the response of greater motor units (Wakeling and Horn 2009). The first and the second bands were averaged to compute the RMS from low frequency components of the signals, which would represent the response of smaller motor units (Wakeling and Horn 2009). These analyses were used to compare bilateral activation for a given muscle.

RMS signals were selected from the full data set for each muscle from each cyclist at four instants of the test: 10, 40, 70 and 90% (averaged from 10 to 20%, 40 to 50%, 70 to 80%, and 90 to 100% of the total test time) (Hug et al. 2004). The overall RMS in both legs were normalized to their individual responses at the start of the test (10 to 20%), where minimal fatigue effects are expected in order to reduce between-subjects variability in EMG data. High and low frequency RMS values were normalized by cyclists' overall RMS from each muscle in order to provide a percentage of the overall signal from each frequency content during the test. All signal processing were conducted using custom made scripts in MATLAB® (Mathworks Inc., Natick, Massachusetts, USA).

Statistical analysis

Data were averaged for the twelve cyclists for the overall, high and low frequency bands. The use of inferential statistics has become controversial during the last years, particularly with growing evidence that shows a disconnection between statistical significance and clinically meaningful changes (Knudson 2009). Therefore, each pair of time-window (i.e. 40-50% vs. 70-80%, 40-50% vs. 90-100%, and so on) was compared using students t-tests for unequal distribution for high, low and overall frequencies for each muscle taken at each lower limb. Effects sizes of each pair of comparison were then scaled for small (ES < 0.20), moderate (ES > 0.20 - 0.8) or large effects (ES > 0.8) (Cohen 1988). Statistical significance was defined when p<0.05 and effect sizes were greater than 0.8. T-tests and effect sizes were computed using a custom made spreadsheet in Excel (Microsoft Inc., USA).

Results

No asymmetries were found among the assessed muscles (Table 2). For that reason, results are shown for the preferred limb. Increase in muscle activation throughout the test was observed for the overall activation of vastus lateralis and biceps femoris, but not for the gastrocnemius medialis (see Figure 1). Low frequency bands from biceps femoris activation were higher at the 90% (60±9% of overall activation) in comparison to 10% (45±16% of overall activation, p = 0.01 and ES = 1.2) and 40% (47±12% of overall activation, p = 0.01 and ES = 1.17) of the total time without changes in high frequency components (see Figure 1).

Differences between muscles activation were analyzed (Table 3). The overall and low frequency content largely differed between gastrocnemius medialis compared to vastus lateralis and biceps femoris. High frequency content followed a similar path with additional differences between biceps femoris and vastus lateralis.

Discussion

We examined the combined effects of fatigue and workload on different frequency components of bilateral lower limb muscle activation. The main findings were that muscle activation increased with workload/fatigue due to greater contribution from overall frequency components in vastus lateralis and biceps femoris, and greater contribution from low (but not from high frequency components) in biceps femoris. Gastrocnemius medialis activation was not influenced by workload/fatigue for any of its frequency bands. These results suggest that in order to sustain exercise workload/fatigue muscle activation may be similarly affected by a large variety of motor units. The combined effect of workload and fatigue elicited during the incremental cycling test to exhaustion led to consistent recruitment during the test in order to sustain maximal use of knee flexors and extensors for greater power production. One disadvantage of the incremental load test used in this study was that it combines effects of fatigue workload, which could be difficult to distinguish when assessing physiological and biomechanical parameters. However, during outdoor cycling and laboratorial tests, cyclists are submitted to a combination of increases in fatigue state and workload (Abbiss et al. 2006; Blake and Wakeling 2012), which lends support to the analyzes of muscle recruitment at the referred protocol.

De Luca (1997) proposed a fatigue mechanism based on increases in the number of small and medium size motor units recruited and a decrease in the recruitment of large size motor units towards exhaustion at submaximal load. Diefenthaler et al. (2012a) showed large decreases in high frequency component of muscle activation during cycling at constant load to exhaustion without changes in low frequency components. Low frequency components of EMG may provide information on the temporal behavior of force fluctuation during steady contractions (Yoshitake and Shinohara 2013a) as well as oscillations in the motor units discharge rate (Yoshitake and Shinohara 2013b). We found increases in overall activation from vastus lateralis and biceps femoris compared to unchanged activation observed in constant load tests to exhaustion (Diefenthaeler et al. 2012a). It is indeed important to highlight that biceps femoris could be differently affected given its reduced cross section area (compared to the vastii group) and to its biarticular role as hip extensor and knee flexor. Further studies could assess if the rectus femoris could be similarly affected given its similar role compared to the hamstrings (i.e. hip to knee joint energy transfer) (Van Ingen Schenau et al. 1994). In line with that, only increases in low frequency contents of biceps femoris activation without changes in high frequency components were observed, in contrast with our previous hypothesis. These results suggest a different physiological adaptation to fatigue expanding the mechanism described by De Luca (1997). The reason for that contrast would be that graded increases in workload from sub maximal to maximal aerobic capacity would allow skeletal muscle to gather energy primarily from aerobic sources (via increases in low frequency components for biceps femoris). These hypotheses should be further assessed by combining local muscle energy analyses (e.g. using near infrared measurements).

Activation of biceps femoris and vastus lateralis were mostly similarly affected by the incremented workload. Previous studies described consistent results only for vastus lateralis (Hug et al. 2003a; Von Tscharner 2009). Evidences suggest that antagonist muscles activation is reduced with fatigue, which would be linked to strategies to minimize co-contraction and improve pedaling effectiveness during maximal sprint cycling (Hautier et al. 2000). However, our findings of greater overall and low frequency activation for biceps femoris towards the end of the test suggest that trained cyclists may use hamstrings to improve pulling forces on the pedal and crank torque during graded exercise testing (Bini and Diefenthaeler 2010; Diefenthaeler et al. 2012b). Biceps femoris was primarily affected by workload/fatigue with earlier increases in low frequency components compared to vastus lateralis in the present study. That could be due to its lower percentage of type I fibers and small size motor units (Staron et al. 2000) An earlier increase in activation may be an attempt to improve force production due to a reduced number of fatigue-resistive fibers, compared to vastus lateralis. Likewise, biceps femoris is a fusiform muscle with less fascicle in parallel (i.e. reduced cross section area) compared to quadriceps muscles (e.g. vastus medialis), which limits its force production (Arnold et al. 2010). Differently, vastus lateralis of cyclists have been shown to present a greater proportion of type I fibers and small size (i.e. more efficient) motor units (Coyle et al. 1991). This scenario enables vastus lateralis to endure longer than biceps femoris and to postpone any changes in muscle activation frequency components. Along with that, biceps femoris acts along with gluteus maximus and other hamstrings in hip extension (Dorel et al. 2009). Therefore increases in activation of biceps femoris could also be due to larger hip joint power production towards maximal workload (Elmer et al. 2011).

The unchanged activation of gastrocnemius medialis may be related to its large contribution to force transfer (not force production) during pedaling action (Fregly and Zajac 1996). Sanderson et al. (2006) observed that gastrocnemius medialis works eccentrically at the recovery phase of crank cycle, differently from other lower limb muscles. We could then infer that to increase force transfer from the limbs to the cranks via eccentric contraction, enhanced gastrocnemius activation may not be required due to greater contribution of non-contractile components of muscle-skeletal system (i.e. fascias and tendons). It was also observed that the high-frequency component of this muscle is greater than vastus lateralis and biceps femoris. It may suggest that gastrocnemius medialis could present a greater proportion of larger motor units (Moritani et al. 1991), compared to biceps femoris (Dahmane et al. 2005) and vastus lateralis.

Studies assessing motor unit recruitment pattern using frequency bands analysis suggest that individuals with varying proportion of fiber-types may have different recruitment patterns adapting to fatigue (Wakeling 2009). Diefenthaler et.al. (2012a) suggested that cyclists use different strategies while activating their muscles during pedaling. In future studies, cyclists could be grouped based on their similarities in muscle activation pattern and on their fiber type distribution, if possible.

The limitations from our study were mostly related to the use of surface EMG to capture muscle activation signals (i.e. adipose tissue filtering effect, cross-talking recording from nearby muscles and muscle temperature in frequency contents of EMG signals). Mathematical correction for differences between muscle in subcutaneous adipose tissue could be an avenue for assessment of differences in frequency content between upper and lower leg muscles. This correction could indicate if differences are due to motor unit profile or to distance from muscle fibers to skin electrodes. Future studies would also apply the frequency band analyses to other lower limb muscles that we were unable to assess.

In conclusion, the present study demonstrated that combined effects from fatigue and workload during an incremental cycling test to exhaustion affected biceps femoris and vastus lateralis muscle activation without changes in gastrocnemius medialis. The main manifestations of fatigue/workload were an increase in the overall activation (for vastus lateralis and biceps femoris) and low-frequency component (only for biceps femoris). These changes may be attributed to a proportion of the muscle fibers and anatomical characteristics of biceps femoris muscle (i.e. bi-articular attachment). Finally, the present study also demonstrated greater high-frequency content in gastrocnemius medialis activation, which highlights a different role of this muscle during cycling

Practical applications

Incremental cycling test led to an increase in activation of main knee joint flexors and extensors but not in plantar flexors during cycling. Biceps femoris changes its recruitment profile due to increases in low frequency content. Changes in pedaling technique could then be assessed for each cyclist via assessments of muscle activation in order to highlight potential needs for supportive training (e.g. strength training).

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