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Research article

Limitations of Spectral Electromyogramic Analysis to Determine the Onset of Neuromuscular Fatigue Threshold during Incremental Ergometer Cycling

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Abstract
Recently, a new method has been proposed to detect the onset of neuromuscular fatigue during an incremental cycling test by assessing the changes in spectral electromyographic (sEMG) frequencies within individual exercise periods of the test. The method consists on determining the highest power output that can be sustained without a significant decrease in spectral frequencies. This study evaluated the validity of the new approach by assessing the changes in spectral indicators both throughout the whole test and within individual exercise periods of the test. Fourteen cyclists performed incremental cycle ergometer rides to exhaustion with bipolar surface EMG signals recorded from the vastus lateralis. The mean and median frequencies (Fmean and Fmedian, respectively) of the sEMG power spectrum were calculated. The main findings were: (1) Examination of spectral indicators within individual exercise periods of the test showed that neither Fmean nor Fmedian decreased significantly during the last (most fatiguing) exercise periods. (2) Examination of the whole incremental test showed that the behaviour of Fmean and Fmedian with increasing power output was highly inconsistent and varied greatly among subjects. (3) Over the whole incremental test, half of the participants exhibited a positive relation between spectral indicators and workload, whereas the other half demonstrated the opposite behavior. Collectively, these findings indicate that spectral sEMG indexes do not provide a reliable measure of the fatigue state of the muscle during an incremental cycling test. Moreover, it is concluded that it is not possible to determine the onset of neuromuscular fatigue during an incremental cycling test by examining spectral indicators within individual exercise periods of the test.

Key words: Cycling, neuromuscular fatigue, surface electromyography, motor unit recruitment, spectral EMG analysis.

Introduction

Lindström and Magnusson (1977) were the first to propose a mathematical model which describes the power spectrum of the surface electromyogram (sEMG) in terms of the conduction velocity of muscles fibres. The main conclusion of their work, namely, that "changes in conduction velocity are always observed as translations of the spectrum along the frequency axis", led to the widespread assumption that an almost perfect linear correlation should exist between spectral sEMG characteristics and conduction velocity (Stulen and De Luca, 1981; Broman et al., 1985; Merletti et al., 1990). This has led numerous authors to investigate the usefulness of spectral sEMG parameters (such as the mean frequency, Fmean, and median frequency, Fmedian) to assess the development of muscle fatigue during cycling (Bouissou et al., 1989; Gamet et al., 1993; Jansen et al., 1997). Indeed, debate still exists about the reliability of spectral sEMG indicators to identify the threshold that demarcates fatiguing from non-fatiguing exercise during an incremental workout on a cycle ergometer (Camic et al., 2010; Hug et al., 2004; Tenan et al., 2011).

Spectral analysis of the surface EMG has yielded conflicting results in studies of dynamic exercise, such as cycling (Bouissou et al., 1989; Gamet et al., 1993; Jansen et al., 1997; Tenan et al., 2011; Wang et al., 2015; Viitasalo et al., 1985). These contradictory findings have often been attributed to the non-stationary character of the sEMG signals during dynamic tasks (Tenan et al., 2011). However, the issue of signal non-stationarity can be largely overcome by using appropriate signal processing methods (Bonato et al., 2001; Karlsson et al., 2000). Therefore, the critical limitation in the spectral analysis of the surface EMG is not related to the stationarity of the signal; but rather the limitation is intrinsic to the generation of surface EMG potentials (Farina et al., 2002). The main confounding factor in sEMG spectral analysis is the distance-dependent filtering effect introduced by the muscle volume conductor: namely, that the contribution of a given motor unit to the spectrum depends on the distance from this unit to the recording electrodes (Dimitrova, 1973). The implication is that a newly recruited motor unit may contribute to the low- or high-frequency region of the spectrum irrespective of its conduction velocity.

Recently, several studies examining the changes in conduction velocity during an incremental cycling test have found that conduction velocity increases with increasing workload up to a maximum value, which coincides with the occurrence of the ventilatory threshold (T-vent), and, subsequently, declines slightly in the transition between T-vent and V'O_2max (Farina et al., 2007; Lenti et al., 2010; Sbriccoli et al., 2009). These results were interpreted to indicate that progressive recruitment of larger motor units occurs from the onset of the incremental test up to approximately T-vent. The later decline in conduction velocity might be the result of de-recruitment of motor units or, alternatively, an expression of the developing muscle fatigue. Therefore, if spectral sEMG parameters were linearly related to conduction velocity, then we would expect a progressive increase in Fmean

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and Fmedian until T-vent, followed by a decrease in these parameters. However, conduction velocity has been shown to depend on many factors such as the external force developed, the instantaneous knee angular speed, and the average pedal rate (Farina et al., 2004b). Therefore, it appears pertinent to re-examine the changes in spectral indicators throughout the incremental test.

The fatigue-induced decreases in spectral indicators and conduction velocity have generally been attributed to the accumulation of metabolic by-products of muscular contraction, especially the extracellular [K+] (Fortune and Lowery, 2007; Juel 1988; van Dieën et al., 2009). Recently, Camic et al. (2010) proposed a method to detect the onset of neuromuscular fatigue during an incremental cycling test by assessing the changes in spectral sEMG parameters within each of the exercise periods of the test. Specifically, the method consists on determining the highest power output that can be sustained without a significant decrease in sEMG Fmean. Theoretically, during the fatiguing power outputs, the increasing levels of extracellular [K+] would result in a significant decrease of conduction velocity (Fortune and Lowery, 2007), which would cause a decrease in spectral indicators. This method, however, has not been validated as yet. Moreover, considering all the potential factors that can influence spectral sEMG indicators, it would be necessary to verify whether, within each of the fatiguing exercise periods of the test, an actual decline in these indicators is found.

The aims of the current study were: (1) to assess the changes in spectral sEMG indicators during the whole duration of the incremental test, and (2) to quantify the changes in spectral parameters within individual exercise periods of the test. The present study was designed to examine the reliability of spectral sEMG indexes to identify the onset of neuromuscular fatigue by analysing the changes in these spectral indexes both during the whole duration of the test and within individual exercise periods. The current work will contribute to clarify whether examination of the changes in spectral indicators within individual workbouts of the incremental test provides a reliable measure of the fatigue state of the muscle, as recently proposed by Camic and colleagues (2010).

Methods

Subjects

Fourteen male semi-professional cyclists volunteered to participate in the study. Their anthropometric and physical characteristics are given in Table 1. Although the sample size may be a bit low, the fact that the sample chosen was homogenous in terms of age, sex, and training level (see below) increased the confidence in the experimental findings. The study was conducted in accordance with the Declaration of Helsinki, and was approved by the Ethics Committee of the Public University of Navarra. Written informed consent was obtained from all participants before inclusion. Participants were asked not to take part in vigorous physical activity for 2 days prior to their test date.

The participants were road cyclists engaged in regular training and amateur road races. Cyclists had a national competitive experience of 3.9 (1.6) years and had performed an average of 18,300 km riding (range 15,000–23,000 km) during the last season. On average, all cyclists trained at least four times a week covering a weekly distance ranging between 400 and 600 km, plus competition or Sunday training. None of the subjects reported any injuries or pathologies of limb muscles or joints.

Screening session

Cyclists underwent a blood test screening prior to participation to check for anemia and possible infections. Blood samples were collected by antecubital venipuncture with Vacutainer system. Red blood cell, white blood cell, platelets, hemoglobin, and hematocrit were determined on a Coulter Counter (model MAX-M). Serum biochemical parameters (glucose, urea, uric acid, creatine kinase, creatin kinase, lactate dehydrogenase, aspartate transaminase, alanine transaminase, aldolase, total proteins, cholesterol and electrolytes) were measured using coupled enzyme reactions on an automatic autoanalyzer (Hitachi 917, Japan). Subsequently, participants were asked to attend an orientation session to become familiarized with the testing apparatus and procedures. All tests were performed on a custom-made cycle ergometer. The saddle and pedals were installed on the cycle ergometer. The participants were required to bring in the saddle, pedals, and cycling shoes from their normal road bicycle. A submaximal incremental test was then performed to familiarize the cyclists with the experimental protocol.

Maximal cycle ergometer test

Participants performed an incremental test to exhaustion on a SRM powermeter (science SRM, SRM GmbH, Germany). The SRM unit consisted of a potentiometer (Powermeter V Science Road Version) connected to a recording system (Powercontrol V). Analysis Software was SRM Training System. Pedal cadence was maintained at 70 rev·min^{-1} during the test, similar to that used in previous studies (Lucia et al., 1999). Seat height and seat setback were adjusted jointly to allow cyclists to adopt their most comfortable cycling posture. Before the incremental exercise test started, cyclists performed 5 min of unloaded cycling. The test was initiated at a workload of 125 W and the load was increased by 25 W every 1 min until the subjects could no longer continue to exercise despite verbal encouragement. At this time, the power achieved was referred to as the maximal power. Heart rate was monitored telemetrically using a Polar Heart Watch system (Polar 610 Plus, Polar Electro Oy, Kempele, Finland).

Analysis of expired gas and determination of ventilatory thresholds (T-vent and RCP)

During the incremental exercise, breath by breath analysis was performed using a turbine flow-meter connected to a face mask (dead space: 30 ml). A side pore of the face mask was connected to fast-response differential paramagnetic oxygen and infrared carbon dioxide analyzers. Throughout the incremental test, the software (Oxycon PRO, Carefusion, Germany) averaged, for 5 consecutive seconds, data of oxygen uptake (VO_2) and carbon dioxide production (VCO_2) and ventilatory parameters, as well as the ventilatory equivalents for O_2 ($EqO_2 = VE/VO_2$) and CO_2 ($EqCO_2 = VE/VCO_2$). The ventilatory threshold (T-vent) was determined using the criteria of an increase in the ventilatory equivalent for oxygen (VE/VO_2) with no increase in the ventilatory equivalent for carbon dioxide (VE/VCO_2) and the departure from linearity of VE (Davis, 1985). The respiratory compensation point (RCP) corresponded to the minimal work rate at which the increase in VE/VO_2 was accompanied by a parallel increase of VE/VCO_2 (Wassermann and McIlroy, 1964). A test-retest reliability study was conducted for the purpose of determining the stability of the performance of our participants. This method was used to determine the intraclass correlation coefficient (ICC). The repeat study was performed on a second day on ten participants (those who lived close to the laboratory where experiments were conducted). Test–retest reliability for VO_{2peak} yielded an intraclass correlation coefficient (ICC) of R = 0.95, with no significant differences between test and re-test values.

Electromyography

Surface electrodes arranged in bipolar configuration were placed on the dominant leg over the vastus lateralis muscle. The electrodes were circular Ag/AgCl electrodes (Kendall Meditrace 100, Tyco, Canada) with a recording diameter of 10 mm and an inter-electrode distance of 20 mm (measured from the nearest lateral borders). The electrodes were positioned according to SENIAM (Surface EMG for Non-Invasive Assessment of Muscles) guidelines (Hermens et al., 1999). Before electrode placement, the skin was adequately prepared (light abrasion with sandpaper and cleansing with rubbing alcohol). The electrodes and cables were secure with surgical tape and cloth wrap to avoid movement-induced artefacts. Surface EMG signals were recorded using MP150 equipment (BIOPAC, Goleta, CA, USA). Raw sEMG signals were amplified with a bandwidth frequency ranging from 10 to 1000 Hz, and digitized online at a sampling frequency of 1000 Hz using the analog-to-digital conversion system of MP150.

Determination of the mean power frequency fatigue threshold

The mean power frequency fatigue threshold (MPF_{FT}) was calculated using the method proposed by Camic et al. (2010). This method is briefly described as follows. During each 1-min workbout of the incremental test, consecutive sEMG epochs (each epoch representing a 128-ms interval chosen during the active period of the vastus lateralis in a single pedaling cycle) were recorded. The first 15-s interval of each 1-min exercise period were not considered since, at the beginning of each period, the cyclist made slight postural adjustments in order to match the target power output and to maintain the required pedal cadence. For each power output of the test, the Fmean value of each of the epochs were calculated and represented as a function of time (see Figure 4 for an example). Then, we identified the lowest power output that resulted in a *significant* negative slope coefficient ($p < 0.05$ in a single-tailed t test) for the sEMG Fmean vs. time relation and the highest power output that gave rise to a *non-significant* slope coefficient ($p > 0.05$). The Fmean fatigue threshold was determined by averaging the above-mentioned power outputs.

Data analysis

Data were first analysed with a commercially-available software (AcqKnowledge, BIOPAC Systems, Goleta, CA, USA) to monitor for any abnormality in sEMG traces. Subsequently, data were exported to Matlab (version R2012b; The Math-Works, Natick, MA, USA) for quantitative analysis using a number of custom scripts. The sEMG signals were filtered using a digital bandpass filter (fourth-order Butterworth) between 15 and 1000 Hz.

Fmean and Fmedian were used as indicators of the power spectrum of the sEMG signal. The spectral analysis was performed over a 128-ms window, as performed in Rodriguez-Falces et al. (2015a). A fast Fourier transformation of 512 points (Hamming window processing) was applied on this window length. Fmean and Fmedian were calculated according to the definition of Stulen and DeLuca (1981). The sEMG epoch for each pedal thrust (descending phase of the pedaling cycle) was chosen from the sEMG bursts, i.e., when the vastus lateralis was active. Each sEMG epoch corresponded to a fixed part of a single pedaling cycle, as performed by Dimitrov et al. (2006). Hence, for each exercise period of the test, 52 values of Fmean (and Fmedian) were calculated (i.e., one for every pedal cycle during 45s at 70 rev·min^{-1}) and analyzed as a function of time.

Statistical analyses

For the evaluation of the changes in spectral sEMG variables throughout the entire test, linear regression analysis was performed and the linear regression coefficients were calculated (to determine the sign and significance of the changes in Fmean and Fmedian). The relation between spectral sEMG indicators and time was also investigated during the interval between the onset of exercise and the occurrence of T-vent and also during the interval between the occurrences of T-vent and the maximal power. For the evaluation of the changes in Fmean during each exercise period of the test, the slope coefficient for each constant workload was calculated from the Fmean vs time relation. To determine whether this slope coefficient was significant, linear regression analysis was performed. Student's paired t-test was used to determine whether there were significant differences in average power output between T-vent and RCP. Significance was set at $p < 0.05$. Data were presented as mean ± SD in the tables.

Results

Physical characteristics and ventilatory fatigue thresholds of cyclists

The anthropometric and physical characteristics of the cyclists involved in the study are shown in Table 1. This table also includes the average values of T-vent and RCP calculated from the incremental test. Note that some of the physiological variables shown in Table 1, such as heart rate, were measured at the end of the incremental test. The power output corresponding to RCP was significantly higher than that of T-vent (370 ± 35 vs. 329 ± 28, $p < 0.05$).

Table 1. Anthropometric (mean ± SD) characteristics, and physiological parameters of the participants (n = 14).

	Mean ± SD	Range
Age (years)	21.2 ± 2.4	18.1 – 27.6
Height (m)	1.79 ± 0.03	1.68 – 1.88
Body weight (Kg)	68.5 ± 4.6	61.2 – 75.9
T-vent (L·min^{-1})	3.2 ± 0.6	1.8 – 4.1
RCP (L·min^{-1})	4.4 ± 0.6	3.2 – 5.4
Heart rate (beats·min^{-1})	195.2 ± 6.9	178 – 203
VO_2max (L·min^{-1})	4.9 ± 0.5	4.2 – 5.6
Maximal Power (W)	399.6 ± 32.1	325 – 450

Changes in spectral sEMG indicators during the incremental cycling test

The time-course of spectral sEMG indicators during the incremental test varied greatly among subjects. This can be appreciated in Figure 1(a), where the values of Fmedian are plotted against time (workload) for individual subjects. Essentially, we identified two different types of behaviour of spectral indicators vs. time, as described next. In the first type, spectral indicators showed an increasing trend during exercise. Regression analysis for the Fmedian-time relation yielded linear regression coefficients between 0.1 and 0.9 Hz·min^{-1} [see histogram of Figure 1(b)]. Similar values were obtained for the Fmean-time relation. This behaviour was observed in half of the participants analysed. For the other half of the participants, Fmedian and Fmean showed a decreasing trend over the course of the incremental test. Linear regression coefficients for the relation between Fmedian and time ranged between -0.3 and -0.9 Hz·min^{-1} [see Figure 1(b)], whereas those for the Fmean-time relation were between -0.1 and -0.9 Hz·min^{-1}. In addition, we observed that the changes in spectral parameters with increasing power output test did not show a consistent behaviour (i.e., they did not change consistently in one direction throughout the incremental test).

To investigate the relation between spectral sEMG indicators and time from the onset of the exercise until the power output corresponding to the ventilatory threshold (where full motor unit recruitment is theoretically reached), the time-courses of Fmedian are plotted up to T-vent for individual subjects [Figure 2(a)]. As can be seen in Figure 2(c), for half of the subjects Fmedian increased up to T-vent, whereas a decreasing trend was observed for the other half. Similar results were observed for Fmean.

The behaviour of spectral sEMG indicators during the last (fatiguing) stages of the incremental test was analysed by assessing the changes in Fmedian and Fmean from the power output corresponding to T-vent to the highest power output that could be maintained, as shown in Figure 2(b). In this figure it can be seen that Fmedian changed slightly for workloads higher than T-vent [this is corroborated by the low correlation coefficients in Figure 2(d)]. Moreover, there were a higher number of subjects showing a positive Fmedian-time relation (9 of 14) from T-vent to maximal power output.

Changes in spectral sEMG indicators within individual exercise periods of the incremental test

Figure 3 shows a representative example of the time-course of power output (a) and Fmean (b) during the incremental test in one subject. At the bottom of these plots, the regression lines of the Fmean vs. time relation are shown separately for the highest power outputs of the test (c). In this example, the regression lines corresponding to 300, 325, and 350 W were practically horizontal, whereas the lines corresponding to 375, 400, and 425 W exhibited a low positive slope. Analysis of individual workouts revealed that, for 9 of 14 subjects, there was a predominance of positive slope coefficients of the Fmean vs. time relation within the last (fatiguing) exercise periods of the test (Table 2). Moreover, for the five subjects for whom there was a predominance of negative Fmedian-time relations, the associated slope coefficients did not reach statistical significance (Table 2).

Relation between ventilatory threshold and spectral sEMG indicators

The possible influence of ventilatory variables on spectral

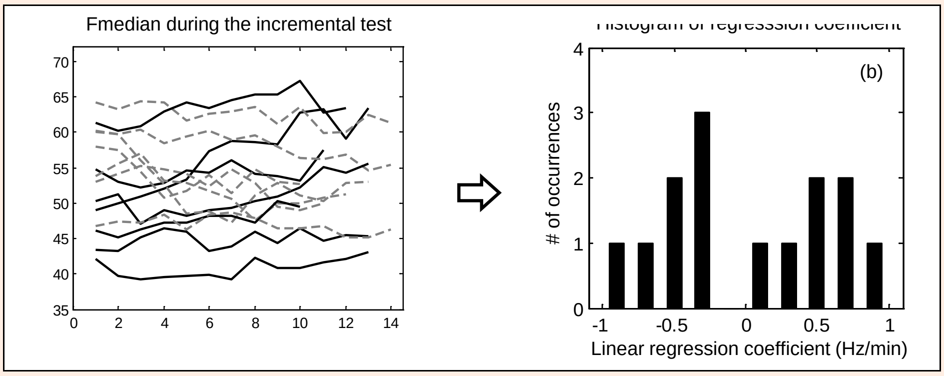

Figure 1. (a) Lines representing the changes in median frequency (Fmedian) over the course of the incremental cycling test for 14 subjects. Black solid lines represent the subjects for which Fmedian showed an increasing trend during exercise, whereas grey dashed lines denote the subjects who showed a decreasing trend during the cycling workout. (b) Histograms of the linear regression coefficients obtained from the relations Fmedian vs time.

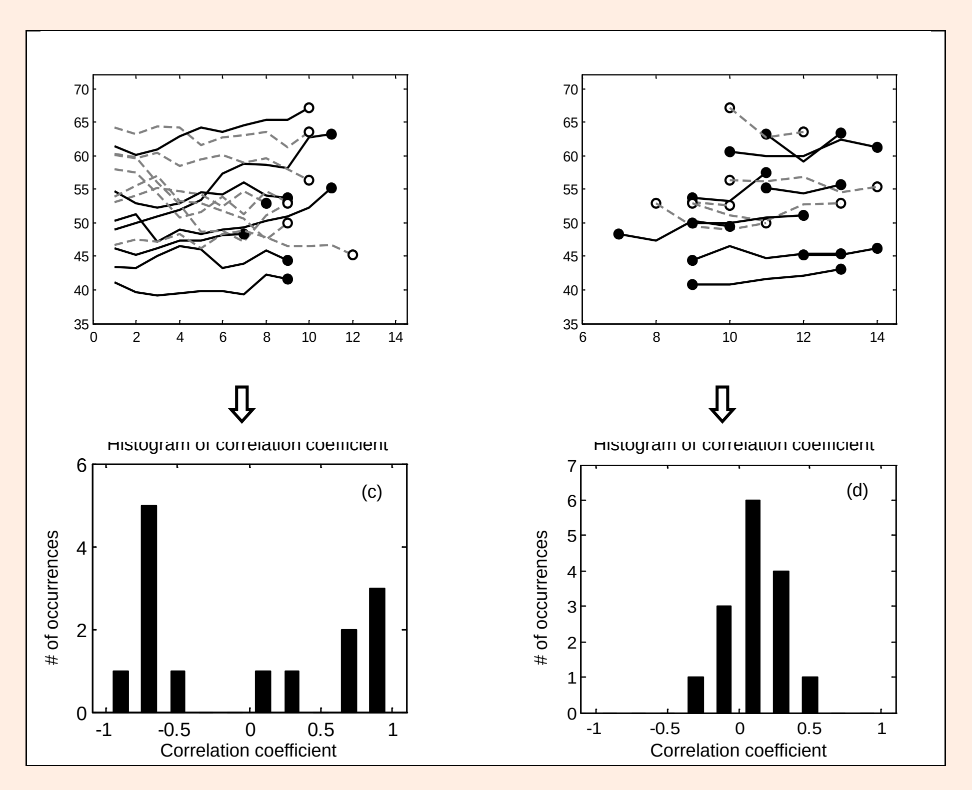

Figure 2. Upper panel - Lines representing the changes in median frequency (Fmedian) from the onset of the incremental cycling test until the power output corresponding to the ventilatory threshold (T-vent) (a) and the changes from T-vent to the maximum power output (b). Black solid lines represent the subjects for which Fmedian showed an increasing trend, whereas grey dashed lines denote the subjects who showed a decreasing trend. Bottom panel - Histograms of the Pearson correlation coefficients obtained from the relations between Fmedian and time for the intervals defined in (a) and (b).

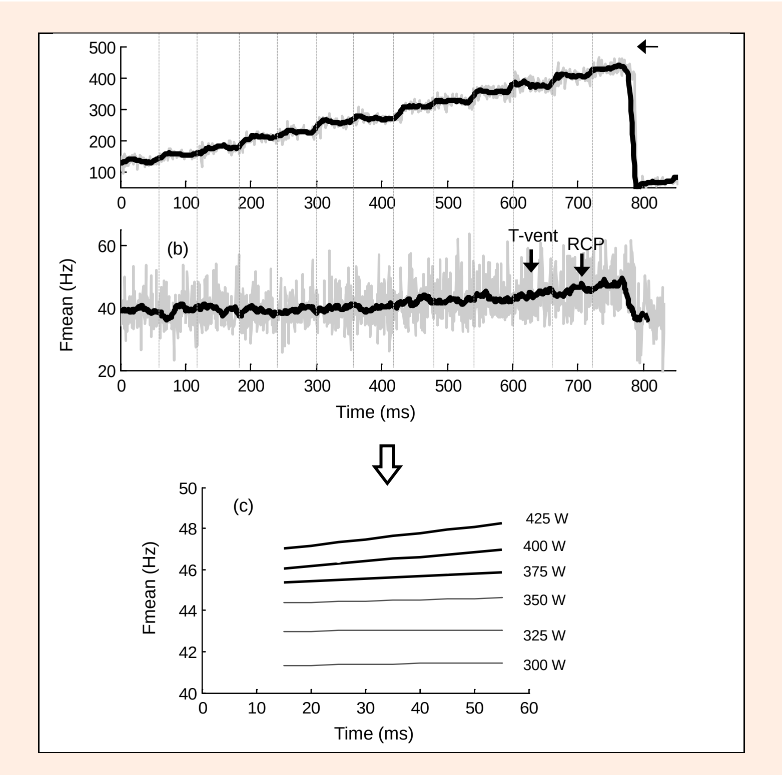

Figure 3. Time plots of power output (a) and mean frequency (Fmean) (b) obtained in one cyclist from the vastus lateralis during the incremental test. Fmean increased slowly from the onset of the protocol until the end of the exercise. In (a) and (b) the black solid lines represent the power and Fmean averaged every 15 bursts. Arrows indicate occurrence of the ventilatory threshold (T-vent), and respiratory compensatory point (RCP). (c) Regression lines corresponding to the Fmean vs. time relation of each power output.

Table 2. Slope coefficients corresponding to the last 5 exercise periods of the incremental test (n = 14).

Cyclist	Slope coefficient (Hz/s)				
	5th to last workbout	4th to last workbout	3rd to last workbout	2nd to last workbout	Last work-bout
#1	.069	.032	.088	.017	.025
#2	.186	.155	-.088	.022	.145
#3	-.124	-.025	-.098	-.008	.124
#4	.087	.034	.192 *	.219 *	.257 *
#5	.017	.108	.207	.028	-.123
#6	-.088	-.032	.067	-.007	-.013
#7	.172	.096	.034	.221 *	.187 *
#8	-.033	.129	.038	-.007	.016
#9	.114	-.126	-.178	-.002	-.014
#10	.093	.139	.144	-.011	.006
#11	.066	-.057	-.124	-.183	-.083
#12	.069	.032	.088	-.017	.025
#13	-.097	-.062	-.143	.008	-.013
#14	-.014	.239 *	.135	.116	.192 *

* significant (positive or negative) slope, as determined by linear regression analysis.

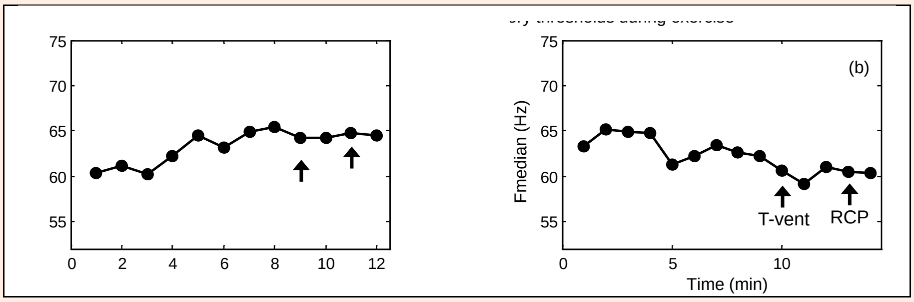

Figure 4. **Time-course of median frequency (Fmedian) and occurrence of ventilatory thresholds in a subject for which Fmedian has an increasing trend during exercise (a) and in a subject for which Fmedian showed a decreasing trend (b). In neither case was Fmedian influenced by the occurrence of the ventilatory threshold (T-vent), the respiratory compensatory point (RCP).**

sEMG indicators was also investigated. Figure 4 shows the time-course of Fmedian and the occurrence of ventilatory thresholds for two subjects. In Figure 4 it can be seen that Fmedian did not show any abrupt change or breakpoint in correspondence with T-vent and/or RCP. In fact, for none of the subjects did the time-course of Fmeand and/or Fmedian exhibit a sudden change coinciding with T-vent and RCP.

Discussion

The main findings of the present study were: (1) Examination of spectral indicators within individual exercise periods of the test showed that neither Fmean nor Fmedian decreased significantly during the last (most fatiguing) exercise periods; (2) Examination over the whole incremental test revealed that half of the participants had a positive relation between spectral indicators and power output, whereas the other had the opposite behaviour; (3) The behaviour of Fmean and Fmedian during the incremental test exhibited a high heterogeneity among individuals.

Heterogeneity and inconsistent behaviour of spectral sEMG indicators during the incremental test

Our results revealed that the behaviour of Fmean and Fmedian with increasing exercise intensity varied greatly among subjects, with approximately half of the participants showing a positive relation between spectral indicators and workload and the other half showing the opposite behaviour. Such high heterogeneity among subjects has already been reported by other authors (Gamet et al., 1993; Jansen et al., 1997; Wang et al., 2015). In addition, spectral sEMG indicators did not change consistently in one direction throughout the incremental test, but rather, in many instances, Fmedian changed in an apparently random manner, without a clear trend. This irregular pattern in the voluntary sEMG power spectrum has been reported even during isometric contractions of increasing strength (Rodriguez-Falces and Place, 2015a).

The inconsistent behaviour and heterogeneity of spectral indicators with increasing exercise intensity can be attributed to a number of factors, the first and most important being the distorting effect introduced by the muscle volume conductor on the sEMG power spectrum. This distortion is generated because the contribution of a given motor unit to the spectrum depends on the distance from this unit to the surface electrodes (Lindström and Magnusson, 1977). As a result, the contribution of a deep motor unit is confined to the lower range of the spectrum, whereas that of a superficial unit is spread over a much wider frequency range (Farina et al., 2002; 2004a). As an example, due to this distance-dependent effect, the recruitment of a slow conduction-velocity motor unit located superficially in the muscle could result in an increase in Fmean and Fmedian, contrary to what is expected. The implication is that in scenarios where there is a progressive recruitment of motor units (such as during an incremental cycling test), because the newly activated units are located at different distances from the recording electrodes, the changes in Fmedian and Fmean resulting from this successive recruitment would be impossible to predict since these changes depend not only on the type of motor unit (slow or fast), but also on the depth of the motor unit in the muscle (Rodriguez-Falces et al., 2015b). The observed inter-individual differences would be partly due to the fact that volume conductor characteristics (thickness and conductivity of the muscle, fat, and skin layers, etc) vary from one subject to another.

Another factor that accounts for the irregular behaviour of sEMG Fmedian with power output is the fact that the conduction velocities of the two main motor unit types are not clearly distinct; rather they have continuous distributions which overlap to a large extent (Troni et al., 1983). This is due to the relative lack of distinction in muscle fiber diameters across motor units with different recruitment thresholds. As a result, recruitment of progressively larger motor units does not result in a systematic increase in spectral indicators. Other factors that could also contribute to the inconsistent behaviour of spectral indices and MCFV are crosstalk and cancellation. In the vastus lateralis, it could well be that, as exercise intensity increases, crosstalk signals from nearby muscles contaminate the target sEMG signal, thereby blurring the expected increase in Fmedian with workload. Cancellation of sEMG amplitude occurs before the interference signal is rectified and it depends on many aspects, including the level of excitation, discharge rate, duration of action potentials, and conduction velocity (Farina et al., 2008).

Interestingly, Gamet et al. (1993) conjectured that the inter-individual differences in Fmedian profiles arise from the different possibilities of balance between various physiological parameters. For example, an increase in Fmedian would occur so long as the effect of motor unit recruitment is not counterbalanced by the accumulation of metabolic by-products. On the contrary, a decreasing trend in Fmedian during the test would be observed in subjects who experience early and marked changes in the concentration of extracellular potassium. However, the recent simulation study of Farina et al. (2002) demonstrate that changes in spectral sEMG parameters during the recruitment phase are not caused solely by alterations in physiological variables. In fact, Farina and colleagues demonstrated that the volume conductor effect (see above) and some anatomical aspects of the muscle (such as the thickness of the subcutaneous layer) can have a great influence on the sEMG power. Nevertheless, whether the volume conductor effect alone can be responsible for the decline in spectral indices observed in some participants from the onset of the test is uncertain.

Inconsistent behaviour of spectral sEMG indicators from exercise onset until ventilatory threshold

Recent studies showed that, during an incremental cycling exercise, conduction velocity increases with increasing workload up to the power output corresponding to T-vent (Sbriccoli et al., 2009; Lenti et al., 2010). The authors interpreted the rise in conduction velocity as evidence that motor units were recruited progressively up to a point of full recruitment, which coincided with T-vent. In order to test to what extent Fmean and Fmedian increase with the recruitment of additional motor units, the present study investigated whether these spectral indices were positively correlated with exercise intensity until T-vent. We found that, in general, Fmean and Fmedian did not increase systematically during the first part of the cycling test. In fact, Fmean and Fmedian showed a positive dependence on power output for only half of the subjects. Moreover, the decrease in spectral indicators observed in the other half of the participants does not seem accidental and has been reported previously (Gamet et al., 1993; Jansen et al., 1997). Collectively, these results suggest that, changes in spectral sEMG indicators are not caused solely by alterations in conduction velocity (as explained above), in agreement with recent findings (Merlo et al., 2005; Farina et al. 2007).

Inconsistent behaviour of spectral sEMG indicators from ventilatory threshold until volitional exhaustion

The limited value of spectral sEMG variables as indicators of fatigue during the incremental test is evidenced by the observation that, for most participants, Fmean and Fmedian either increase or remained stable during the higher fatiguing power outputs (i.e., between T-vent and maximal power output), whereas conduction velocity has been reported to slightly decrease at these high intensities (Lenti et al., 2010; Sbriccoli et al., 2009). The increase in spectral indexes is surprising since, during the last stages of the test, participants reach a critical level of fatigue, as witnessed by the high levels of interstitial [K+] reported (Hug et al. 2003; Tenan et al., 2011). The absence of decrease in Fmean and Fmedian during the last part of the incremental exercise is in agreement with the theory that changes in spectral sEMG indicators are not sensitive to alterations in plasma [K+] and lactate concentration (Jansen et al., 1997; Tenan et al., 2011). It is precisely the increased interstitial [K+] the prevailing explanation for the decrease in conduction velocity (Fortune and Lowery, 2007; Juel et al., 1988). The lack of correspondence between spectral indices and conduction velocity is due to the fact that Fmean and Fmedian are global parameters of the surface EMG signal, which are affected by a number of factors (such as the distorting effect of the muscle volume conductor, crosstalk, and cancellation) other than the metabolic changes of the muscle.

Changes in spectral sEMG indicators within individual exercise period of the incremental test

Recently Camic and colleagues (2010) proposed that, during an incremental cycling workout, the onset of neuromuscular fatigue could be determined by examining the changes in spectral sEMG parameters within the individual exercise periods of the test. Specifically, the method searches for the highest power output that can be maintained during a certain interval (exercise period) without a significant decrease in Fmean and Fmedian. Therefore, the method is based on the assumption that, when fatigue reaches a critical level, a marked decline in sEMG spectral parameters should be observed within the corresponding exercise period. However, this assumption has been challenged by the present findings. First, we have found that, for 9 of 14 participants, there were positive slope coefficients for the Fmean vs. time relationship in the last fatiguing periods (see Figure 3). Moreover, for the cyclists for whom the Fmean vs. time relationship yielded a negative slope coefficient, this coeffient was not significant. Therefore, our results questioned the usefulness of the sEMG frequency-based method proposed by Camic et al. (2010) to detect the threshold of neuromuscular fatigue

The absence of a significant decrease in Fmean and Fmedian during the last (fatiguing) exercise periods of the incremental test is intriguing and deserves further consideration. One plausible explanation is that full motor unit recruitment is actually not reached at T-vent, but rather recruitment continues for higher power outputs. This explanation is consistent with the idea that, during a progressive cycling test, “neuromuscular fatigue” of the vastus lateralis, as assessed by the sEMG frequency characteristics, is not a significant limiting factor in performance (Tenan et al., 2011). This means that participants may reach volitional exhaustion before the vastus lateralis achieves a critical level of “neuromuscular fatigue” (that would result in a significant decline in Fmedian), thereby suggesting that termination of exercise occurs due to cardiovascular limitations. Another factor that may counteract the expected decrease in Fmean and Fmedian produced by fatigue is muscle shortening. Reduction of muscle fibres’ length has been shown to shift the power spectrum to higher frequencies (Inbar et al., 1987; Schulte et al., 2004). Therefore, as the exercise intensity increases, the progressive shortening of the leg muscles would tend to increase spectral parameters. Finally, muscle temperature is expected to increase over the course of the incremental test, thereby exerting a positive influence on spectral parameters (Gamet et al., 1993). It seems reasonable to assume that the above processes (muscle fatigue, motor unit recruitment, muscle shortening, and increasing temperature) are initiated when contractile activity is started, and that they coexist during practically the whole duration of the ergometer exercise. The coexistence of these processes would make it difficult to quantify either process independently. Moreover, the lack of a distinct decline in spectral sEMG indicators (and also in conduction velocity) at the end of the test is probably due to the fact that muscle fatigue and the aforementioned factors act on the sEMG power spectrum in different directions.

Relation between spectral sEMG indicators and ventilatory thresholds

In the current study the time-course of Fmean and Fmedian did not show any abrupt change or breakpoint in correspondence with T-vent and RCP, indicating that changes in ventilatory variables do not influence the power spectrum of the sEMG signal. This result is interesting as it contradicts a recent study by Camic et al. (2010) who found significant positive correlations between spectral sEMG variables and RCP. To interpret such result, the authors hypothesized that the changes in spectral characteristics and in ventilation parameters should be related to a common physiological mechanism, such as the accumulation of interstitial [K+]. However, the lack of interaction between spectral parameters and ventilatory fatigue thresholds reported here suggests that respiratory centers and sEMG characteristics are probably related to different mechanisms of fatigue.

Conclusion

In summary, this study shows that the behaviour of spectral sEMG indicators during an incremental cycling test is highly inconsistent and varies greatly among individuals. Such heterogeneous and irregular behaviour is most likely due to several factors influencing the generation of the surface EMG signal, which includes the distance-dependent effect of the muscle volume conductor, cross-talk, cancellation, muscle length, temperature, and the lack of distinction in fibre diameter across motor units with different recruitment thresholds. It has been shown that spectral sEMG indexes did not decrease significantly within the last fatiguing exercise periods of the incremental test, as recently proposed. In agreement, spectral indexes did not show a global decrease in the transition between the ventilatory threshold and the maximal power output. Collectively, the above findings indicate that spectral sEMG frequencies, assessed both during the whole test and during individual exercise periods, are not reliable measures of the fatigue state of the muscle during cycling and, consequently, they do not allow identifying the onset of neuromuscular fatigue in an incremental cycling exercise.

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Key points

- The behaviour of spectral EMG indicators during the incremental test exhibited a high heterogeneity among individuals, with approximately half of the participants showing a positive relation between spectral indicators and workload and the other half showing the opposite behaviour.
- None of the spectral EMG indicators examined (Fmean nor Fmedian) decreased significantly between the ventilatory threshold and the highest power output.
- Examination of spectral indicators within individual exercise periods of the test showed that neither Fmean nor Fmedian decreased significantly during the last (most fatiguing) exercise periods.

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