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- 1 Title: Insights gained into the interpretation of surface electromyograms from the gastrocnemius
- 2 muscles: a simulation study.
- 3 **Authors:** Luca Mesin¹, Roberto Merletti² and Taian M.M. Vieira^{2,3}
- 4 1 Department of Electronics, Politecnico di Torino, Torino, Italy.
- 5 2 Laboratory for Engineering of the Neuromuscular System, Politecnico di Torino, Torino, Italy.
- 6 3 School of Physical Education and Sports, Federal University of Rio de Janeiro, Rio de Janeiro,
- 7 Brazil.
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- 9 Word count: 3247
- 10 Corresponding author:
- 11 Taian Vieira
- 12 Dipartimento di Elettronica, Politecnico di Torino
- 13 Corso Duca degli Abruzzi 24, Torino, 10129 ITALY
- 14 Tel: 0039-011-4330476
- 15 Fax: 0039-0114330404
- 16 e-mail: <u>taian.vieira@delen.polito.it</u>
- 17

1 ABSTRACT

2 Interpretation of surface electromyograms (EMG) is usually based on the assumption that the surface representation of action potentials does not change during their propagation. This 3 assumption does not hold for muscles whose fibers are oblique to the skin. Consequently, the 4 interpretation of surface EMGs recorded from pinnate muscles unlikely prompts from current 5 knowledge. Here we present a complete analytical model that supports the interpretation of 6 experimental EMGs detected from muscles with oblique architecture. EMGs were recorded from 7 8 the medial gastrocnemius muscle during voluntary and electrically elicited contractions. Preliminary indications obtained from simulated and experimental signals concern the spatial 9 10 localization of surface potentials and the myoelectric fatigue. Specifically, the spatial distribution of surface EMGs was localized about the fibers superficial extremity. Strikingly, this localization 11 increased with the pinnation angle, both for the simulated EMGs and the recorded M-waves. 12 13 Moreover, the average rectified value (ARV) and the mean frequency (MNF) of interference EMGs increased and decreased with fatigue, respectively. Furthermore, the degree of variation in ARV and 14 MNF did not depend on the pinnation angle simulated. Similar variations were observed for the 15 experimental EMGs, although being less evident for a higher fiber inclination. These results are 16 discussed on a physiological context, highlighting the relevance of the model proposed here for the 17 18 interpretation of gastrocnemius EMGs and for conceiving future experiments on muscles with pinnate geometry. 19

1 1. INTRODUCTION

The modeling of surface electromyograms (EMGs) has been sought for the interpretation of experimental data (Dimitrova and Dimitrov 2003; Merletti et al., 1999b; Roeleveld et al., 1997), for the development of algorithms aimed at information extraction (Duchene and Hogrel, 2000; Mesin et al., 2009), and for didactic purposes (Merletti et al., 1999a). Available models for the generation of surface EMGs rely both on numerical (Lowery et al., 2004; Mesin et al., 2006) and analytical (Block et al., 2002; Farina et al., 2004b;Gootzen et al., 1991; Mesin, 2006) approaches.

8

The assumption that the volume conductor is invariant in the direction of propagation of 9 10 intracellular action potentials allowed for the development of fast analytical models for the simulation of surface EMGs (Farina and Merletti, 2001; Farina et al., 2004a). If a volume conductor 11 is space invariant, the surface representation of a motor unit action potential does not change with 12 its propagation. Such space invariance is frequently assumed when simulating, processing and 13 interpreting experimental EMGs (Lindstrom and Magnusson, 1977; Reucher et al., 1987). 14 Therefore, much of the insights gained into the interpretations of surface EMGs from the use of 15 mathematical models are valid, exclusively, for muscles whose geometry fits in the assumption of 16 space invariance. 17

18

Some of the muscles investigated by surface EMG cannot be approximated assuming the muscle 19 fibers to be parallel to the skin. The fibers of the gastrocnemius muscle, for example, are inclined 20 with respect to the skin surface and extend from the deep to the superficial aponeurosis (Kawakami 21 et al, 1998; Narici et al., 1996). Surface electrodes positioned on the calf are, thus, located above the 22 superficial aponeurosis, where muscle fibers attach. Considering that action potentials propagate 23 along the muscle fibers, their propagation along the oblique gastrocnemius fibers contributes to the 24 surface EMGs with a component toward muscle extremities and another toward the skin/tibia. 25 Therefore, the area on the skin surface upon which the action potential of a single muscle fiber 26

distributes likely depends on how inclined the muscle fibers are (i.e. its pinnation angle) (Vieira et
al., 2011). Currently existing models do not provide indications of how the surface EMG relates to
the pinnation angle. Which physiologically relevant information might be extracted from surface
EMGs in the pinnate gastrocnemius muscle is unknown.

5

In this study we simulate a pinnate muscle, with fibers inclined in the depth direction, to interpret 6 surface EMGs detected from the gastrocnemius muscles. A complete mathematical model, which 7 includes muscle fibers with finite length and three layers of tissues, is presented here for the 8 generation of single fiber action potentials (SFAP). In particular, we simulate the distribution of 9 10 motor unit action potentials (MUAPs) on the skin and the interference EMG for different degrees of inclination of muscle fibers. The implications for the interpretation of how action potentials 11 distribute on the skin surface and for the estimation of muscle fatigue are addressed as well. 12 Ultrasound images and experimental EMGs are recorded from the human medial gastrocnemius 13 (MG) muscle to investigate how much theoretical and empirical data correspond. 14

15

16 **2. METHODS**

17 2.1 Mathematical model

The simulation model was developed by extending our previous work (Mesin and Farina, 2004), which considered the impulse response of a two layer volume conductor. Three layers were considered here (skin, fat and muscle; figure 1). Moreover a complete model (including finitelength fibers) was implemented to generate single fiber action potentials (SFAP).

22

Figure 1

Libraries of MUAPs were generated from the simulated SFAPs. These libraries were used with a model of spatial and temporal recruitment of motor units (MUs) to simulate interference EMGs during fatiguing contractions.

1 Full details on the model and the simulated EMGs are described in the Appendix.

2

3 2.2 Experimental signals

Single-differential EMGs were recorded from the medial gastrocnemius (MG) muscle to test for the correctness of the information gained from simulated signals. In particular, we investigated how the amplitude of surface potentials varies with the pinnation angle. This is of remarkable interest since this variation likely provides information of how localized the gastrocnemius activity might be. Moreover, we were interested in understanding whether experimental and simulated EMGs in pinnate muscles are comparable during fatiguing contractions.

10

Three male subjects (age: 33, 29, 27years; body mass: 78, 80, 75kg; height: 182, 178, 180cm)
participated in two protocols designed to compare experimental and simulated signals.

13

Protocol 1: Sixteen surface electrodes (10 mm IED) were used to record EMGs during electrical 14 stimulation. Two ankle angles were considered; foot in neutral position ($\sim 20^{\circ}$ pinnation angle) and 15 plantar flexed (~35° pinnation angle). With an adhesive pre-gelled electrode (cathode; figure 6a) 16 placed carefully on the leg, bipolar current pulses were delivered at 2 pps and for 20s to the tibial 17 18 posterior nerve. The anode electrode (80×50 mm; soaked cloth) was positioned immediately above the patella with elastic Velcro straps. Stimulation amplitude was as minimal as possible to allow for 19 the detection of the firstly observable M-wave. Low stimulation amplitude was chosen to recruit the 20 least number of MUs and, thus, to better isolate the effect of ankle angle on the distribution of M-21 waves amplitude on the skin. Averaged M-waves were obtained after triggering the 15 single-22 differential EMGs. 23

24

Protocol 2: EMGs were recorded with an array of eight electrodes (5mm interelectrode distance –
IED) when the subjects exerted isometric plantar flexion at 60% MVC. The contraction lasted for

1 30s to ensure the occurrence of myoelectric manifestation of fatigue. Fatigue plots were created 2 from the average rectified value (ARV) and the mean frequency (MNF) calculated on 500ms epochs (i.e. both descriptors are plotted with respect to their initial values). ARV and MNF indices 3 were calculated as indicated in Merletti et al. 1990. This protocol was applied once with the foot 4 dorsal flexed (pinnation angle $\sim 10^{\circ}$) and once in neutral position (pinnation angle $\sim 20^{\circ}$), with 5 min 5 interval between trials. Plantar flexion torque was measured and displayed to the subjects. Subjects 6 7 were in prone position and their feet were firmly secured to a footplate, with the lateral malleolus 8 being coaxial to the centre of rotation of the torque meter. MVC values were determined for each pinnation angle as the maximum torque measured across three attempts separated by 5 minutes. 9

10

EMGs were amplified (gain ranged from 1k to 5k; 10-500 Hz EMG-USB amplifier, LISiN and 11 OTBioelettronica, Turin), and sampled at 2048 Hz with a 12bit A/D converter (±2.5V dynamic 12 13 range). Ultrasound images were taken with a linear probe (3.86 cm long; Fukuda Denshi, UF 4000, 7.5 MHz) and pinnation angles were estimated with the precision of one degree. A custom-made 14 neuromuscular stimulator (LISiN, Turin), equipped with a hybrid output stage, was used in the first 15 protocol. In the second protocol, load cells output was amplified (150Nm full-scale amplifier; 16 MISOII, OTBioelettronica, Turin) and then converted to values of ankle torque. After cleansing of 17 18 the skin with abrasive paste and water, ultrasound scanning was used to place electrodes on the MG muscle. Specifically, electrodes were positioned above the sheath of aponeurotic tissue and parallel 19 to the surface projection of the pinnate MG fascicles. Care was taken to ensure that the array and 20 the ultrasound probe had similar orientation, which provided an ultrasound image with as many 21 fascicles as possible. 22

23

24 **3. RESULTS**

25 **3.1 Preliminary Simulations**

Some preliminary simulations are shown in figures 2 and 3, in order to compare the surface potentials produced by fibers with different arrangements (parallel or slightly inclined with respect to the skin). In particular, the potential distribution over the skin surface is considered for fixed time samples in figure 2, whereas the signals detected as functions of time from arrays of electrodes sampling the potential in specific points aligned to the fibers are shown in figure 3.

6

Simulations showed that the amplitude and the shape of surface potentials change with the obliquity 7 of muscle fibers (figure 2). When simulating fibers parallel to the skin, the surface distribution of 8 action potentials was symmetric with respect to the innervation zone (IZ). Conversely, by 9 10 increasing the fiber inclination, the surface potential became more asymmetric, as the contribution of the deeper source was attenuated and more diffused than that of the most superficial source. The 11 root mean square (RMS) difference between the surface distribution of potentials simulated for 10° 12 and those computed for the other pinnation angles $(0^{\circ}, 5^{\circ}, 15^{\circ}, and 20^{\circ})$ is shown as a function of 13 the source position (figure 2b). As expected, greater RMS differences were obtained for higher 14 variations between angles. The greatest difference occurred when the source was close to the 15 extinction region. 16

17

18 Regardless of whether simulating monopolar or single-differential signals, variations in the amplitude of surface potentials across electrodes depended on the pinnation angle. By simulating an 19 electrode array placed parallel to the x-z projection of a superficial muscle fiber with 0° pinnation 20 angle, both monopolar and differential potentials appeared with equal amplitudes on either sides 21 from the IZ (left panel in figure 3a). For deep fibers, only the standing waves produced by the 22 generation and extinction of simulated potentials are seen in the EMGs, respectively (right panel in 23 figure 3a). By inclining muscle fibers, the amplitude distribution of surface potentials concentrated 24 progressively more toward the superficial tendon (left panels in figure 3b,c). Interestingly, pinnate 25 fibers simulated 10cm away from the electrodes contributed to the surface EMGs with potentials of 26

markedly smaller amplitude (3-5 times smaller; right panels in figure 3b,c). Therefore, surface
electrodes on pinnate muscles sample only from sources located nearby and nowhere else.

3

The inclination of muscle fibers affects also the shape and the propagation of surface potentials. 4 Close inspection of figure 3 reveals that the surface potential from fibers parallel to the skin has the 5 same duration, wherever it is sampled on the skin. Propagation of these potentials is also evident in 6 the figure. Potentials from pinnate fibers undergo large shape variations when recorded from 7 different channels, so that conduction velocity (CV) cannot be estimated properly (Farina and 8 Merletti, 2004). Specifically, they have shorter duration when detected closer to the superficial 9 tendon. Moreover, the delay between successive peaks or valleys of potentials simulated for 10 adjacent channels is not the same for pinnate fibers. Then, surface EMGs detected from muscles 11 with fibers not parallel to the skin unlikely show the propagation of action potentials along muscle 12 13 fibers.

14

Figures 2 and 3

15

16 **3.2 Amplitude distribution of EMGs**

In figure 4, simulated single fiber potentials are investigated to quantify how much the spatial 17 18 distribution of surface EMGs changes as a function of the pinnation angle. Surface potentials were compared for volume conductors with various pinnation angles, from 0° to 45°, and for two 19 thicknesses of the fat layer. Contour plots of simulated surface potentials are shown at a given time 20 (figure 4b). To quantify the diffusion of the surface potential, the median and the first and third 21 quartiles were investigated along orthogonal sections of contour plots (figure 4c). The potential 22 generated using a model of parallel fibers was more diffused (interquartile interval: ~40mm) over 23 the skin with respect to that generated with a pinnate geometry (interquartile interval: less than 24 25mm for pinnation angles higher than 10° ; figure 4). 25

As for the simulated SFAPs, the amplitude distribution of electrically elicited EMGs depended on 1 2 the degree of inclination of MG fibers. When MG fascicles were $\sim 20^{\circ}$ oblique, the firstly emerging M-waves appeared on the most proximal muscle portion, from the channel 1 to 5 (figure 5b). The 3 ARV amplitude of M-waves distributed evenly across these channels (interquartile interval: 4 53.7mm). Strikingly, for the same stimulation amplitude and higher pinnation angle (~35°), the 5 distribution of ARV amplitude changed chiefly across channels (interquartile interval: 32.2mm), 6 7 with the most proximal channels detecting the largest M-waves (figure 5c). Additionally, M-waves recorded from the pinnate gastrocnemius muscle showed neither a delay nor a phase opposition 8 between consecutive channels. 9

10

Figure 4 and 5

11 **3.3. Simulated and experimental manifestation of fatigue in surface EMGs**

Simulations indicate that amplitude and spectral descriptors might be used to study the myoelectric 12 manifestations of fatigue in the pinnate gastrocnemius muscle. Single-differential EMGs, simulated 13 and recorded with seven channels aligned to the longitudinal projection of muscle fibers, are shown 14 for two pinnation angles (10° and 20°; figure 6). Both simulated (session 2.4) and experimental 15 signals showed myoelectric manifestation of fatigue, with ARV and MNF increasing and 16 decreasing with time, respectively (fatigue plots; figure 6). Interestingly, changes in amplitude and 17 18 frequency were more variable for the experimental EMGs; and this high variability increases with the pinnation angle (Table 1). 19

20

21 **4. DISCUSSION**

A model for the simulation and interpretation of surface EMGs in pinnate muscles is proposed here. Simulations indicate that the obliquity of muscle fibers has a marked effect on the surface EMG (figure 2). The higher the fibers pinnation the more localized was the surface distribution of action potentials, with electrodes closer to the fibers end detecting remarkably higher potentials. This localized distribution of surface potentials is more evident for single-differential than for monopolar

Figure 6, Table 1

signals, likely due to the higher selectivity of differential derivations. Experimental EMGs from the
MG substantiated the predictions posed by our simulations.

3

While there are architectural differences between skeletal muscles, it seems worthy to ask whether the surface EMGs recorded from pinnate muscles are as informative as those recorded from fusiform muscles. Understanding how the amplitude distribution of surface potentials changes with the pinnation angle and the possibility of using surface EMGs to study muscle fatigue are of particular interest.

9

10 4.1 Localization of surface EMGs depends on the pinnation angle of the gastrocnemius muscle Our simulations revealed that surface electrodes on muscles with fibers not parallel to the skin 11 sample from nearby sources and from nowhere else. This key result is substantiated from our 12 13 observations that: i) surface potentials were localized over the superficial tendon of pinnate fibers, with the main contribution being due to the extinction phase of intracellular action potentials (figure 14 3); ii) although the representation of surface potentials was 50% less localized when trebling the fat 15 thickness, it was still considerably more concentrated on the superficial tendon than the surface 16 representation of potentials in parallel fibers (figure 4); iii) two pinnate fibers with distal tendons 17 separated by 10cm produced surface potentials with separated amplitude distributions. 18 Consequently, the pinnation brings a wider cross section of the muscle into the view of surface 19 electrodes than that available for parallel-fibered muscles. It is worth to mention that the localized 20 distribution shown in figures 3 and 4 refers to the amplitude of SFAPs. As the amplitude of MUAPs 21 corresponds to the algebraic summation of several SFAPs, the localized representation of individual 22 MUAPs in the surface EMGs depends on whether MUs have small territories (Vieira et al., 2011), 23 Here, we are not interested on the size of MUs territories but on how much the amplitude 24 distribution of SFAPs changes with the pinnation angle. 25

Electrical stimulation of the tibial nerve revealed a somewhat similar localization of surface M-1 2 waves detected from the MG muscle (figure 5), with respect to that observed for simulated potentials. The smallest stimulation amplitude leading to the first observable M-waves was chosen 3 to ensure that only a few MUs were stimulated. By keeping constant the stimulation amplitude, the 4 effect of pinnation angle on the localization of M-waves was isolated. M-waves with similar 5 amplitude were observed for the four most proximal channels when stimulation pulses were 6 delivered with the foot in neutral position (figure 5b, $\theta = 20^{\circ}$). With the foot plantar flexed (figure 7 5c, $\theta = 35^{\circ}$), the amplitude of M-waves distributed unevenly across the four most proximal 8 9 channels. Larger M-waves appeared for the more proximal channels. While the variation in ankle 10 joint angle could have induced variation in the stimulation site, as the nerve moves beneath the stimulation electrode, the similitude of EMGs likely indicates that the same population of MUs was 11 stimulated in both foot conditions. 12

13

Surface EMGs convey unique information regarding the activation of the pinnate gastrocnemius 14 muscle. In muscles whose fibers are parallel to the skin, surface electrodes detect the propagation of 15 MUAPs (Farina et al, 2002; Merletti et al., 2003). In the gastrocnemius muscle, the amplitude of 16 surface EMGs varies with the number of active fibers beneath the recording electrodes. Very 17 18 recently, for example, we used the model presented here to validate our estimations of the longitudinal size of MUs territory in the human MG muscle (Vieira et al., 2011). Specifically, we 19 20 observed that with respect to the MG length, the fibers of individual MUs extended for only a short 21 distance (less than 4cm). Then, activation of individual MUs leads to regional activation of the MG 22 muscle. Indeed, the extensive evidence positing localized activation of the calf muscles in humans and cats is not surprising (Eng and Hoffer, 1997; English and Weeks, 1989; McLean and Goudy, 23 24 2004; Staudenmann et al., 2009; Vieira et al., 2010a,b; Wolf et al., 1998). If the nervous system takes advantage of muscle architecture to shape the recruitment of MUs (Kennedy and Cresswell, 25

2001; Vieira et al., 2011), the high-density surface electromyography (Merletti et al., 2010) could
provide a mean to study the regional organization of activity in the pinnate gastrocnemius muscle.

3

4 **4.2** Myoelectric manifestation of fatigue in pinnate muscles

Testing for the possibility of studying fatigue manifestation in skeletal muscles from the surface 5 EMG is not simple. Several factors affect the muscles undergoing fatigue: 1) different strategies of 6 7 MU recruitment and firing rate; 2) variations in load sharing between synergistic muscles, especially for those spanning the same joint as soleus and gastrocnemius (McLean and Goudy, 8 2004); 3) changes in shape of intracellular action potentials (Arabadzhiev et al., 2005); 4) 9 10 synchronization of MUs (Mesin et al., 2009); 5) variable decrease of CV for muscle fibers of different physiological types (Rainoldi et al., 2008). To keep simulations simple, we considered the 11 same percentage of CV changes for all MUs. Additionally, the same spatial and temporal 12 13 recruitment of MUs was simulated for two different pinnation angles, so as to isolate the effect of fibers inclination on surface EMGs. 14

15

Even though it was not possible to observe the changes in CV directly, it was possible to observe 16 myoelectric manifestations of gastrocnemius fatigue in the amplitude and spectral EMG descriptors. 17 18 Because of the muscle oblique architecture, and considering the scattering of IZs, the extinction of action potentials at the superficial aponeurosis contributed chiefly to the surface EMGs simulated 19 (figure 3). As a consequence, electrodes located over the aponeurotic layer do not detect the same 20 potential propagating toward either the superficial or deep aponeurosis. Instead, they record 21 standing waves likely resulting from the end-of-fiber effect (Stegeman et al., 1997). This 22 interpretation is supported by spike-triggered EMGs, which did not show physiological delays 23 between surface potentials detected by consecutive electrodes located upon the superficial 24 aponeurosis (Vieira et al., 2011). Thus, estimation of CV, which showed remarkable properties in 25 the investigation of myoelectric fatigue in parallel-fibered muscles (Mesin et al., 2009), cannot be 26

obtained from the gastrocnemius muscle, at least not upon its superficial aponeurosis. Nevertheless, simulations showed that ARV and MNF of surface EMGs are both affected by variations in CV.

3

Although at different extents, the experimental and simulated manifestations of fatigue were 4 observed in the pinnate gastrocnemius muscle. Simulations did not show any reliance of the 5 variations in ARV and MNF on the pinnation angle. Nevertheless, the variability of ARV and MNF 6 7 were different for experimental data recorded for two pinnation angles (Table 1). This suggests that 8 some factors, which were kept constant in the simulations, were involved in the contraction of the MG muscle at different pinnation angles. It has been shown, for example, that variations in activity 9 10 within and between calf muscles occur when subjects are asked to sustain a low and isometric torque of plantar flexion (McLean and Goudy, 2004; Tamaki et al., 1998). Variable activation 11 within the same gastrocnemius muscle was observed even for different directions of ankle force 12 13 (Staudenmann et al., 2009) and in quiet standing (Vieira et al., 2010a,b). Considering the more localized distribution of surface potentials for the more oblique geometry (figures 4), and the small 14 territories of gastrocnemius MUs (Vieira et al., 2011), surface electrodes seem to provide very 15 selective recordings for high pinnation angles. Therefore, variations in recruitment and firing rate of 16 MUs during a fatiguing contraction would affect more severely the surface EMGs detected from 17 18 more oblique MG fibers.

19

20 **4.3 Future perspectives**

The model presented here opens new perspectives for investigating the calf muscles activation, either by supporting the interpretation of experimental data or by providing theoretical grounds upon which future studies will be designed. The surface distribution of simulated action potentials (figures 3 and 4), for example, supports our prediction that postural activation of the MG muscle is organized regionally for the control of quiet standing posture (Vieira et al., 2010a). Similarly, our simulation results indicate that amplitude and spectral EMG descriptors could be potentially useful
 to study fatigue in the gastrocnemius muscle.

3

4 5. CONCLUSIONS

A new model for the simulation of surface EMGs in pinnate muscles was developed and applied to 5 interpret experimental data obtained from the gastrocnemius muscles. The most striking result was 6 the localized representation of surface potentials. Because of the gastrocnemius pinnate 7 8 architecture, simulated EMGs and experimental M-waves were both detected only by few consecutive electrodes. Higher pinnation angles led to more localized potentials. The regional 9 10 organization of gastrocnemius activity in different motor tasks could, then, be investigated with surface electromyography. The potentiality of the model was also shown for the investigation of 11 myoelectric fatigue with amplitude and spectral indexes. Both descriptors varied during fatigue, 12 13 either for experimental or simulated EMGs. The greater variability in the amplitude and frequency of experimental EMGs, observed for higher pinnation angles, was likely due to different 14 recruitment strategies. 15

16

17 **Conflict of Interest Statement**

18 None of the authors has any conflict of interest concerning the publications of this work.

19

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Table 1: Variability of the amplitude (ARV) and frequency (MNF) descriptors of experimental and simulated surface EMGs, calculated as the standard deviation of residuals resulting from the fitting of a first order polynomial to the observed data (dashed lines in figure 4).

	Experime	ental EMGs	Simulated EMGs	
Pinnation angle (deg)	ARV (µV)	MNF (Hz)	ARV (a.u.)	MNF (Hz)
10	67	4.16	4.21	3.61
20	108	4.68	4.20	3.62

1 FIGURE CAPTIONS

2 Figure 1 a) Schematic representation of the volume conductor model, including the definition of the coordinate system and the geometry of the simulated muscle fibers. The muscle layer is 3 considered as homogeneous and anisotropic, with fibers inclined with respect to the skin surface. 4 Fat and skin layers are homogeneous and isotropic. b) Ultrasound image of the gastrocnemius, 5 showing the pinnation of muscle fibers. c) Longitudinal section of the simulated volume conductor, 6 indicating the notation used for determining the analytical solution. d) Sampling of the 7 phenomenological model of the transmembrane current proposed in Rosenfalck (1969), using 10 8 impulse sources. 9

Figure 2 a) Surface potential generated in muscle fibers with three different pinnation angles (0°, 5°, and 10°). The signals were simulated using the model shown in Figure 1a,c. b) Root mean square (RMS) difference between the EMG map generated for the pinnation angle of 10° and the maps computed for pinnation angles of 0°, 5°, 15°, and 20°. Two different thicknesses of the fat layer were simulated. RMS values are expressed in percentage and as a function of the position of the source (measured as the distance of each of the two transmembrane current sources from the innervation zone).

Figure 3 Simulation of surface EMG signals detected with an electrode array located over the longitudinal projection of a single muscle fiber on the skin surface. Monopolar and singledifferential signals are shown for different fibers, corresponding to different pinnation angles or to different positions.

Figure 4 Simulated (a; see Appendix) and experimental (b; see section 2.2) EMGs are shown for two pinnation angles (10° and 20°) during an isometric fatiguing contraction at 60% of maximal voluntary contraction (MVC). Interference EMGs were simulated with the muscle fiber conduction velocity - CV - decreasing by 1%/s of the initial value. The volume conductors simulated and ultrasound images of the MG muscle are shown on top (arrows indicate detection points). The same firing pattern and population of 340 motor units were simulated for two pinnation angles (10° and 1 20°). A short time epoch of raw signals is depicted in the middle panel. Fatigue plots are shown in 2 the bottom, including averaged rectified values (ARV) and mean frequency (MNF), estimated for 3 epochs of 500 ms duration (n = 60 epochs) and for each pinnation angle. Values were averaged 4 across channels (n = 7 channels). Dashed lines indicate the best linear fitting to ARV and MNF 5 traces, calculated with the least square method.

Figure 5 Spatial distributions of surface potentials as a function of the pinnation angle. a) 6 7 Longitudinal view of the volume conductors considered. When the pinnation angle is higher than zero, one of the tendons is located at the muscle/fat interface, representing the superficial 8 aponeurosis. The case in which fibers are parallel to the skin surface is also considered, for two 9 10 muscle depths. b) Contour plots of the rectified value, averaged over time (ARV), of the simulated surface potential, indicating the median and the first and third quartiles for orthogonal sections 11 crossing the location of maximal amplitude. Note the asymmetry of contours for pinnation angles 12 13 greater than zero; ARV amplitude concentrates close to the fiber end (the superficial aponeurosis). c) Median, first and third quartiles of ARV in the direction longitudinal (x) and transversal (z) to the 14 fibers are shown for different pinnation angles and for two fat layers. 15

Figure 6 Spatial distribution of single-differential M-waves detected from the medial 16 gastrocnemius (MG) muscle at two pinnation angles. a) Array of 16 surface electrodes, positioned 17 18 on the MG muscle, and the stimulation electrode, located above the tibial nerve branch supplying the same muscle. Note that the most proximal EMG electrodes were nearer to the stimulation 19 electrode. b) Raw triggered (black traces) and averaged (gray traces) M-waves detected with the 20 foot positioned so as to result in a pinnation angle of about 20°. The ultrasound image obtained with 21 the foot in this same position is shown on the top. c) the same as in b) for $\sim 35^{\circ}$ pinnation angle. 22 Note that the stimulation artefact was most evident for the most proximal channels, which were the 23 closest to the cathode electrode. 24

- 26

1 Fig 1



3 Fig 2



b) RMS difference (percentage) between EMG maps computed for $\theta = 0^{\circ}$, 5°, 15° and 20° and that obtained for $\theta = 10^{\circ}$



1 Fig 3









2 Fig 5



4 Fig 6

