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Surface electromyography low-frequency content: Assessment in isometric conditions after electrocardiogram cancellation by the Segmented-Beat Modulation Method

Agnese Sbrollini, Annachiara Strazza, Silvia Candelaresi, Ilaria Marcantoni, Micaela Morettini, Sandro Fioretti, Francesco Di Nardo, Laura Burattini*

Department of Information Engineering, Università Politecnica delle Marche, via Brecce Bianche 12, Ancona, 60131, Italy

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ABSTRACT

Background: Surface electromyography (SEMG) is widely used in clinics for assessing muscle functionality. All procedures proposed for noise reduction alter SEMG spectrum, especially in the low-frequency band (below 30 Hz). Indeed, low-frequency band is generally addressed to motion artifacts and electrocardiogram (ECG) interference without any further investigation on the possibility of SEMG having significant spectral content. The aim of the present study was evaluating SEMG frequency content to understand if low-frequency spectral content is negligible or, on the contrary, represents a significant SEMG portion potentially providing relevant clinical information.

Method: Isometric recordings of five muscles (sternocleidomastoideus, erectores spinae at L4, rectus abdominis, rectus femoris and tibialis anterior) were acquired in 10 young healthy voluntary subjects. These recordings were not affected by motion artifacts by construction and were pre-processed by the Segmented-Beat Modulation Method for ECG deletion before performing spectral analysis.

Results: Results indicated that SEMG frequency content is muscle and subject dependent. Overall, the 50^{th}[25^{th};75^{th}] percentiles spectrum median frequency and spectral power below 30 Hz were 74[54; 87] Hz and 18[10; 31] % of total (0–450 Hz) spectral power.

Conclusions: Low-frequency spectral content represents a significant SEMG portion and should not be neglected.

1. Introduction

Surface electromyogram (SEMG) is a noninvasive recording of the electrical activity of muscles during activation, widely used in clinics for assessing their functionality. SEMG is acquired by placing electrodes on the body surface, has a high repeatability and permits a long-term monitoring [1]. Besides SEMG of the muscle of interest, skin electrodes also record other corrupting signals, such as power-line noise, motion artifacts, crosstalk from neighboring muscles and electrocardiogram (ECG). Specifically, ECG interference is high when SEMG electrodes are applied on head or thoracic muscles [2] and tends to decrease going toward limbs. ECG cancellation from SEMG recordings is challenging since ECG and SEMG spectra partially overlap. Indeed, SEMG spectrum is believed to fall below 450 Hz [3] while most ECG spectrum falls below 50 Hz (even though some components may reach 100–200 Hz) [4]. To attenuate ECG and other interferences, SEMG is typically processed using linear high-pass filters; setting of the cut-off frequency, however, is still controversial and mainly ranges from 10 Hz to 30 Hz [5–11]. Despite its broad use, high-pass filtering is rather basic and not very efficient, because always eliminating some frequency components of the signal of interest (that is SEMG) and maintaining some frequency components of the corrupting signal (that is ECG). Consequently, more sophisticated and effective methods have been proposed in literature. Among these, independent component analysis [10,12–15] is a blind source separation technique that aims to instantaneously separate mixed sources from a recording by recognizing fundamental patterns [15]; however, it typically also removes some frequency components of the signal, thus introducing the possibility of signal loss [13]. Adaptive filtering [2,16,17] requires noise-adapting selection of control parameters, which has a big influence on the filter performance so that it is usually not repeatable among studies [16]. Eventually, hybrid wavelet techniques [14] have shown to be efficient in case of marginal ECG

* Corresponding author.
E-mail addresses: a.sbrollini@pm.univpm.it (A. Sbrollini), a.strazza@pm.univpm.it (A. Strazza), s1066375@studenti.univpm.it (S. Candelaresi), i.marcantoni@pm.univpm.it (I. Marcantoni), m.morettini@univpm.it (M. Morettini), s.fioretti@univpm.it (S. Fioretti), f.dinardo@univpm.it (F. Di Nardo), l.burattini@univpm.it (L. Burattini).

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contamination when combined with independent component analysis [12,15], and to introduce errors strictly dependent on user-defined spectral analysis parameters when combined with non-negative matrix factorization algorithms [15].

All proposed filtering procedures to attenuate ECG interference alter SEMG spectrum, especially in the low-frequency spectral content. Indeed, low-frequency spectral components are assumed to be related to noise (motion artifacts and ECG) and thus are usually neglected without any further investigation. However, as far as we know, absence of physiological content related to the low-frequency band has not been shown. Rather, there exist applications, such those relative to motor-unit analysis or to SEMG fatigue effects, where it is necessary to maintain the low-frequency components unaltered [8,18,19]. In order to further investigate additional physiological significance and clinical utility of SEMG low-frequency content, new SEMG filtering procedures able to preserve this spectral band are needed. Studies focusing on SEMG frequency content are quite sporadic and general [14,20,21]. In an interesting paper on ECG deletion from SEMG [14], authors sustain that SEMG low- and high-frequency bands may reveal different aspects of muscle properties. Moreover, a preliminary study from our group [22] on a single muscle (left rectus abdominis) suggested that, after ECG deletion by the Segmented-Beat Modulation Method (SBMM, briefly described in Appendix A) [23,24], SEMG maintains a significant amount of spectral components (up to 20%) in the low-frequency range [22]. Thus, the aim of the present study was evaluating SEMG frequency content, especially in the low-frequency range, to understand if power contributed by the low-frequency harmonics can be considered negligible (as commonly assumed) or, on the contrary, it represents a significant power portion and thus should not be neglected (as occasionally suggested). To this aim, SBMM was applied to remove ECG interference from both simulated as well as real SEMG tracings before performing the spectral analysis. Specifically, the simulation study was performed in order to evaluate if SBMM is able to correctly remove the ECG interference without introducing SEMG distortion. Instead, the electrophysiological study, which involved real SEMG tracings of isometric contractions, was performed to investigate if the SEMG low-frequency components are negligible or not. Isometric contractions were considered to avoid motion artifacts. The electrophysiological study involved 10 young healthy subjects who were asked to perform a movement analogous to that performed during the Functional-Reach test [25–27]. This movement allows to get isometric contractions of both upper and lower body muscles among which sternocleidomastoideus, erectores spinae at L4, rectus abdominis, rectus femoris and tibialis anterior.

2. Materials and methods

2.1. Signal modeling

Any isometric recording (IREC) can be seen as the superimposition of SEMG, which is the signal of interest, and other corrupting signals, mainly ECG and instrumentation noise. Isometry guarantees absence of motion artifacts. Under the assumption of using modern electronic technology (which is substantially immune to instrumental noise [7,28]) that integrates power-line notch filters, IREC can be modeled as the summation of SEMG and ECG:

$$IREC = SEMG + ECG. \quad (1)$$

Thus, Eq. (1) represents an approximation of IREC when all interferences but ECG can be neglected.

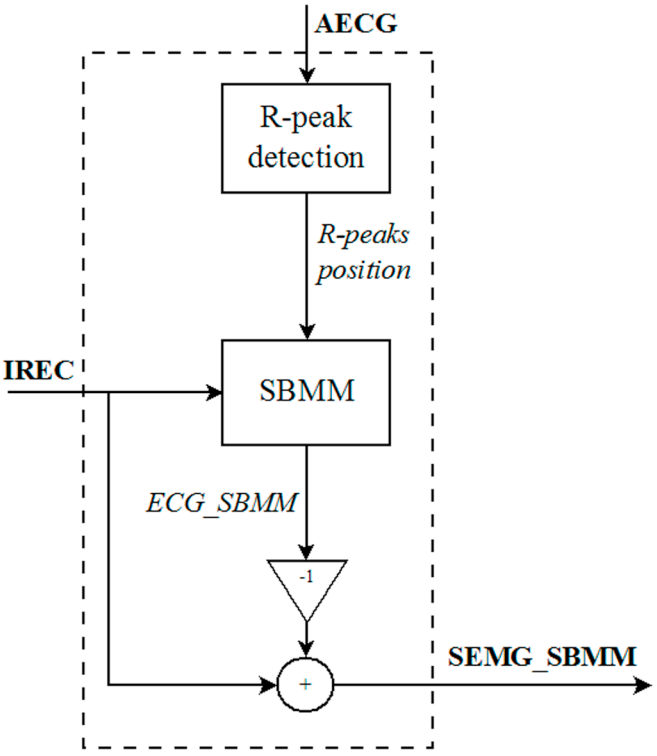

Fig. 1. Segmented-Beat Modulation Method (SBMM) based procedure to get the surface electromyogram (SEMG) from an isometric recording (IREC) by electrocardiographic (ECG) interference subtraction.

2.2. Filtering procedure for electrocardiogram deletion

The filtering procedure proposed here for deleting ECG components from IREC is SBMM-based. Details on SBMM may be found in Refs. [23,24]; a brief description is also reported in Appendix A. SBMM is an algorithm originally proposed for ECG estimation from a noisy recording. In this work, it was innovatively applied to get a clean SEMG from IREC (Fig. 1). Precisely, at first IREC was treated as an ECG recording affected by SEMG (which initially serves as noise) and thus submitted to SBMM to estimate ECG (ECG_SBMM). If IREC does not include ECG, ECG_SBMM is a zero-constant signal. Once obtained, ECG_SBMM can be subtracted from IREC in order to estimate SEMG (SEMG_SBMM, the signal of interest):

$$SEMG\_SBMM = IREC - ECG\_SBMM \quad (2)$$

Use of SBMM in electromyographic applications is possible only if an additional recording, mainly including an electrocardiographic signal (AECG), is simultaneously acquired to IREC. Thus, ECG (that is the electrocardiographic component of IREC, Eq. (1)) and AECG are two morphologically-different but simultaneously-recorded representations of the cardiac electrical activity, and thus are characterized by the same R-peaks time occurrence. AECG is used for identification of R-peaks position, needed to SBMM for ECG_SBMM evaluation (Fig. 1; Appendix A).

2.3. Testing studies

2.3.1. Simulation study

The simulation study was designed to show how the proposed SBMM-based filtering procedure works in controlled conditions and, in

particular, to demonstrate that SBMM is able to remove ECG interference from SEMG recordings without spectral distortions.

To simulate an isometric contraction, the simulated SEMG (SimSEMG) was synthesized as in Ref. [29]. Specifically, SimSEMG was simulated as a bandlimited (0–150 Hz) stochastic process with zero-mean Gaussian distribution (mean: 0.00 mV, standard deviation: 0.20 mV; amplitude: 0.80 mV, that is four times standard deviation) as indicated in Ref. [29]. Sampling frequency was 1000 Hz. Instead, a simulated ECG (SimECG) was obtained as a 40-fold concatenation of a clean, 750 ms long real beat [30]. The electrocardiographic pattern of SimECG contains the P wave, the QRS complex and the T wave, in order to simulate a physiological cardiac behavior. SimSEMG and SimECG were pre-processed to a 10 Hz high-pass filter and to a 50 Hz notch filter for simulating instrumentation filters (see the Electrophysiological study below). The low-pass filter at 500 Hz was avoid because guaranteed by the sampling frequency at 1000 Hz, in according to Nyquist criterion. They were summed to get a simulated IREC (SimIREC):

$$SimIREC = SimSEMG + SimECG. \quad (3)$$

Three simulation cases were considered, termed SIM1, SIM2 and SIM3, in which SimIREC was corrupted by a high, low and no electrocardiographic interference, respectively. Specifically, SimSEMG amplitude was kept constant at 0.80 mV in all cases, while three different SimECG amplitude values (measured as maximum minus minimum of the basic beat) were considered, that are 0.50 mV, 0.25 mV and 0.00 mV (i.e. no SimECG), respectively. Low-amplitude values of SimECG (typically around 1 mV in the standard electrocardiography) were considered to simulate ECG interference of a real SEMG acquisition. SimECG interference was respectively clearly visible, hardly visible and not present in SimIREC. In all cases, SimECG served also as AECG.

2.3.2. Electrophysiological study

The electrophysiological study was designed to evaluate SEMG spectral distribution. Isometric recordings (IREC) were acquired in 10 healthy volunteers (5 males and 5 females; age: 24.0 ± 2.6 year; height: 169.6 ± 10.4 cm; weight: 62.0 ± 11.7 kg; dominant right side in all cases) using the multichannel recording system Step32 (Version PCI-32 ch2.0.1. DV), Medical Technology, Italy, which integrates an analogic 50 Hz notch filter for power-line noise removal. Sampling frequency was originally 2000 Hz, then digitally reduced to 1000 Hz after application of an antialiasing band-pass filter to limit bandwidth at 0–500 Hz [31]; resolution was 12 bits. Single-differential SEMG probes with fixed geometry constituted by box-integrated Ag/Ag–Cl disks (size: $7 \times 27 \times 19$ mm; gain: 1000, high-pass filter: 10 Hz, input impedance > 1.5 GΩ at $f = 0$ Hz, CMRR > 113 dB, input referred noise < 1 μV_{rms}) were used. An interelectrode center-to-center distance of 12 mm was chosen by following the SENIAM (seniam.org) recommendations to avoid crosstalk [20,32]. To assure proper electrode-skin contact, each used electrode was dressed by highly-conductive gel after rubbing skin with abrasive conductive paste so that noise deriving from electrode-skin interference was considered negligible [33].

Electrodes were positioned over the following dominant-side muscles (Fig. 2): sternocleidomastoideus (head), erectores spinae at L4 level (trunk), rectus abdominis (trunk), rectus femoris (leg) and tibialis anterior (leg). Only one channel per muscle was recorded. Under supervision of an expert physiotherapist, electrodes positioning followed the SENIAM recommendations for electrode location and orientation

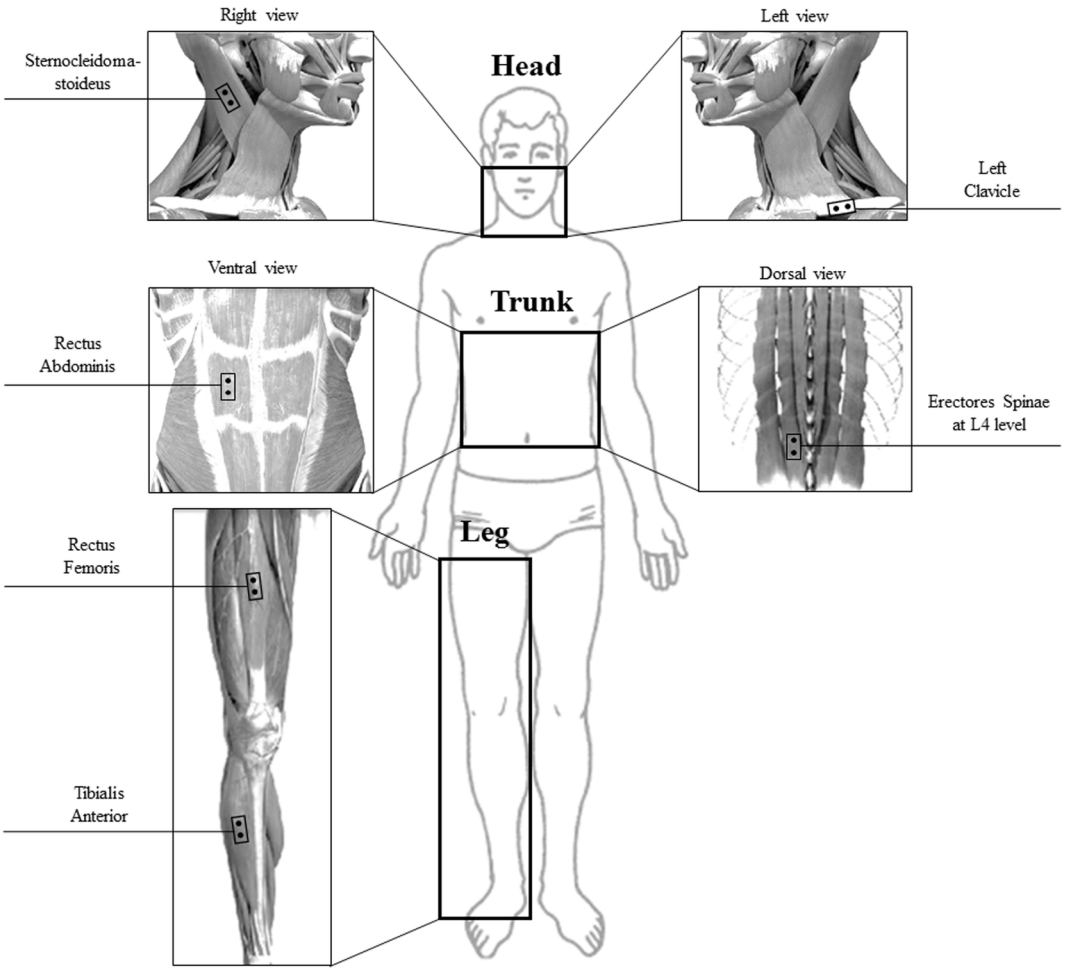

Fig. 2. Electrode positioning setup for isometric recording (IREC) and additional recording (AECG; left clavicle) relative to the electrophysiological study.

over muscle with respect to tendons, motor point position and fibers direction [32]. An additional electrode was attached over the left clavicle, where there is no muscle, to record AECG. Before applying electrodes, skin surface was cleaned, dried, abraded and, if necessary, shaved. To assure proper electrode-skin contact, each electrode was dressed by highly-conductive gel.

After being accurately instructed, the subject was asked to extend the dominant arm as far forward as possible, to keep it perpendicular to the trunk for at least 30 s approaching an isometric contraction, analogously to what done during a Functional-Reach test [25–27]. The extension was repeated three times, since this number was considered as a tradeoff between the need of reducing measure noise (by averaging over repetitions) and the need of not having fatigue effects. Only the IREC 30 s windows acquired during isometric contractions were used for our clinical evaluations.

The present study was undertaken in compliance with the ethical principles of Helsinki Declaration and approved by the institutional expert committee; all participants gave their informed consent prior to testing.

2.4. Spectral analysis and statistics

The signal-to-noise ratio (SNR), defined as in Eq. (4), was used to quantify the amount ECG interference affecting SimIREC and IREC [34]:

$$SNR = 10 \times \log_{10}\left(\sigma^2_{signal} \Big/ \sigma^2_{noise}\right) \quad (4)$$

Specifically, "signal" and "noise" represent the electromyographic and the electrocardiographic components, respectively. When not known, "noise" was estimated using SBMM. SNR = ∞ indicates absence of ECG interference.

Spectral analysis was performed by computing the normalized (by its area) Fourier power spectral density (PSD(f), with f being frequency), estimated via Welch's method (windows length: 1 s). The median frequency (MdnF) was computed together with the percent amount of power (PSD%) in f1 < f ≤ f2 band:

$$PSD\%(f1,\ f2) = \frac{PSD(f1 \div f2)}{PSD(0 \div 450)} \times 100, \quad (5)$$

where PSD(f1 ÷ f2) is the power content between two specific frequencies, f1 and f2 respectively. Thus, PSD(0 ÷ 450) represents the total power, since 0 Hz and 450 Hz are the lowest frequency and the highest frequency characterizing SEMG spectrum, respectively [3]. Two bands were considered: the low-frequency band, defined for f1 = 0 Hz and f2 = 30 Hz; and the main-frequency band, defined for f1 = 30 Hz and f2 = 450 Hz.

Three types of MdnF and PSD%(0,30) variabilities were considered: the inter-muscle variability, described by distributions of these variables over the five muscles of a subject in an acquisition; the inter-subject variability, described by distributions of these variables over the 10 subjects relatively to a specific muscle in an acquisition; and the intra-subject variability, described by distributions of these variables over three acquisitions, relatively to a single muscle of a single subject. Intra-muscle, inter-muscle and inter-subject variabilities among the

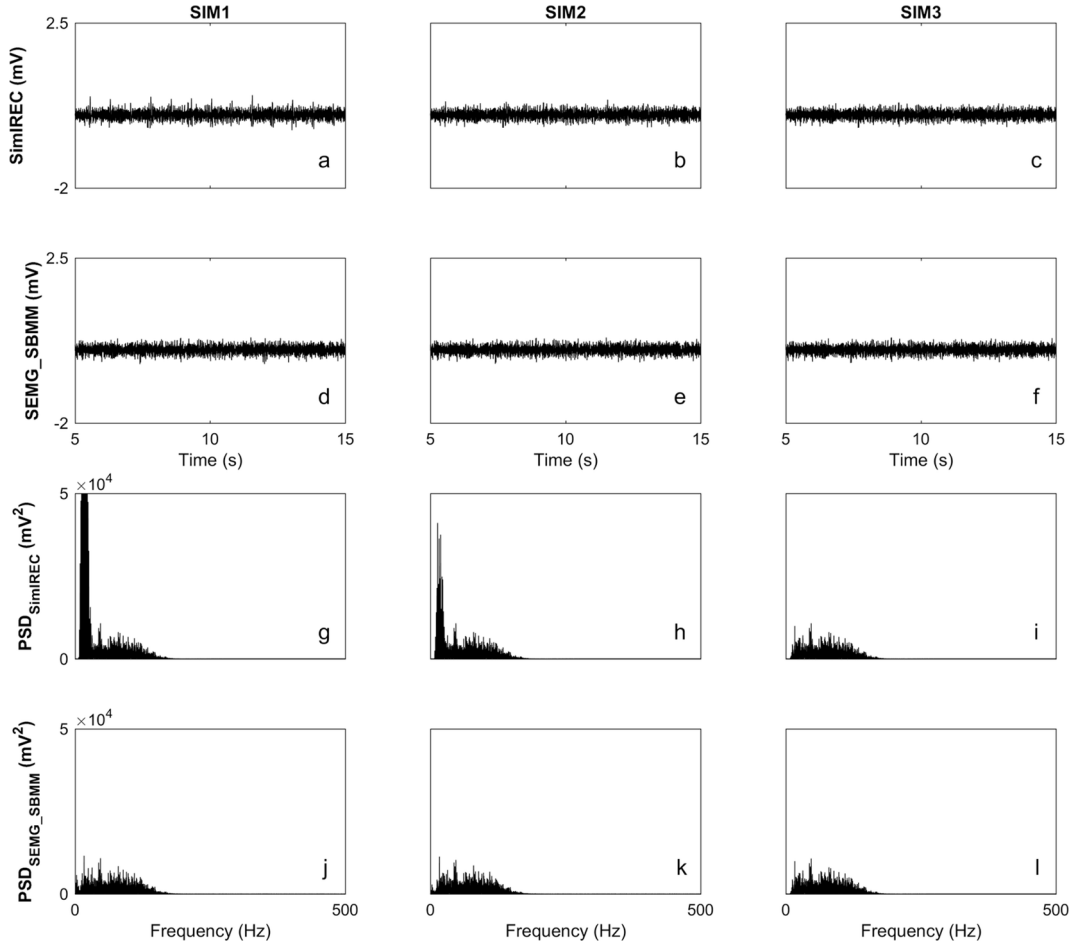

Fig. 3. Qualitative representation of signals involved in the simulation study in both time domain (panels a to f) and frequency domain (panels g to l). Simulated electromyogram estimation by Segmented-Beat Modulation Method (SEMG_SBMM; panel d to f) was obtained after applying Segmented-Beat Modulation Method based filtering procedure to simulated isometric recording (SimIREC; panels a to c). Power Spectrum Density of SimIREC ($PSD_{SimIREC}$; panels g to i) and SEMG_SBMM (PSD_{SEMG_SBMM}; panels j to l) were obtained by Fourier transform.

Table 1
Quantitative characterization of SimECG, SimIREC and SEMG_SBMM relative to the simulation study.

Simulation	Signal	SNR (dB)	MdnF (Hz)	PSD%(0,30)
SIM1	SimSEMG	∞	74	16
	SimIREC	5	57	33
	SEMG_SBMM	∞	73	18
SIM2	SimSEMG	∞	74	16
	SimIREC	10	70	21
	SEMG_SBMM	∞	74	16
SIM3	SimSEMG	∞	74	16
	SimIREC	∞	74	16
	SEMG_SBMM	∞	74	16

MdnF: Median Frequency; PSD: Power Spectrum Density; SEMG_SBMM: Surface ElectroMyoGram estimation by Segmented-Beat Modulation Method; SIM1: First SIMulation case; SIM2: Second SIMulation case; SIM3: Third SIMulation case; SimIREC: Simulated Isometric RECording; SimSEMG: Simulated Surface ElectroMyoGram; SNR: Signal-to-Noise Ratio.

acquisitions were compared using the Wilcoxon ranksum test; statistical significance was set at 0.05.

Finally, we quantified a fatigue factor (FF) in each subject for each muscle to evaluate the fatigue related to repetitions. FF is defined as the percentage of the half of interquartile interval (i.e. the 75th percentile minus the 25th percentile) over the median value of the MdnF. If FF is lower than 15%, the fatigue effects are considered negligible. Variables distributions were described in terms of median (50th percentiles) [25th percentiles; 75th percentiles].

3. Results

3.1. Simulation study

A qualitative representation of signals involved in the simulation study is depicted in Fig. 3, while quantitative results are reported in Table 1. In SIM1 (Fig. 3 left column of panels) SimIREC (Fig. 3a) showed a high (visible) SimECG interference; consequently, its SNR was quite low (SNR = 5 dB; Table 1). In SEMG_SBMM (which is SimSEMG estimated by SBMM when applied to SimIREC; Fig. 3d) SimECG has been filtered out so that its SNR was ∞, as for SimSEMG (Table 1). Analogous results (SNR = ∞ for both SimSEMG and SEMG_SBMM; Table 1) were obtained for SIM2 (Fig. 3b and e) in which, however, SimECG interference was low (SNR = 10 dB; Table 1). Eventually, SNR was equal to ∞ (Table 1) in all SimSEMG, SimIREC (Fig. 3c) and SEMG_SBMM of SIM3 (Fig. 3f), where SimIREC was affected by no interference.

For what concerns frequency analysis (Fig. 3, panels g to l), SimIREC low-frequency components due to ECG interferences (SIM1 and SIM2) were very high (Fig. 3, panels g and h) due to perfect periodicity of SimECG. SimSEMG MdnF was 74 Hz in all simulations (Table 1); SimIREC MdnF increased from 57 Hz in SIM1, to 70 Hz in SIM2, to 74 Hz in SIM3 (Table 1), due to a decreasing amplitude (from 0.50 mV to 0.25 mV–0.00 mV) of SimECG interference; eventually, in all cases, SEMG_SBMM MdnF remained quite stable (73 Hz in SIM1, 74 Hz in SIM2 and 74 Hz in SIM3; Table 1). Moreover, in SimIREC, PSD%(0,30) decreased with decreasing SimECG amplitude (33% in SIM1, 21% in SIM2 and 16% in SIM3; Table 1) whereas, in SEMG_SBMM, it remained quite stable (18% in SIM1, 16% in SIM2 and 16% in SIM3; Table 1). PSD%(30,450) trend over simulation cases was complementary.

3.2. Electrophysiological study

A qualitative representation of signals involved in one acquisition of subject 5 is depicted in Fig. 4. In this example, IREC amplitude (Fig. 4, first row of panels) was different in different muscles and the affecting ECG interference was clearly visible only in IREC of sternocleidomastoideus (Fig. 4a) for which SNR was the lowest (5[1; 11] dB). According to SBMM, ECG interference was also present in IREC of erectores spinae at L4 (Fig. 4b) and rectus abdominis (Fig. 4c), even though not visible (SNR: 10[8; 12] dB and 6[2; 9] dB, respectively), but not in IREC of

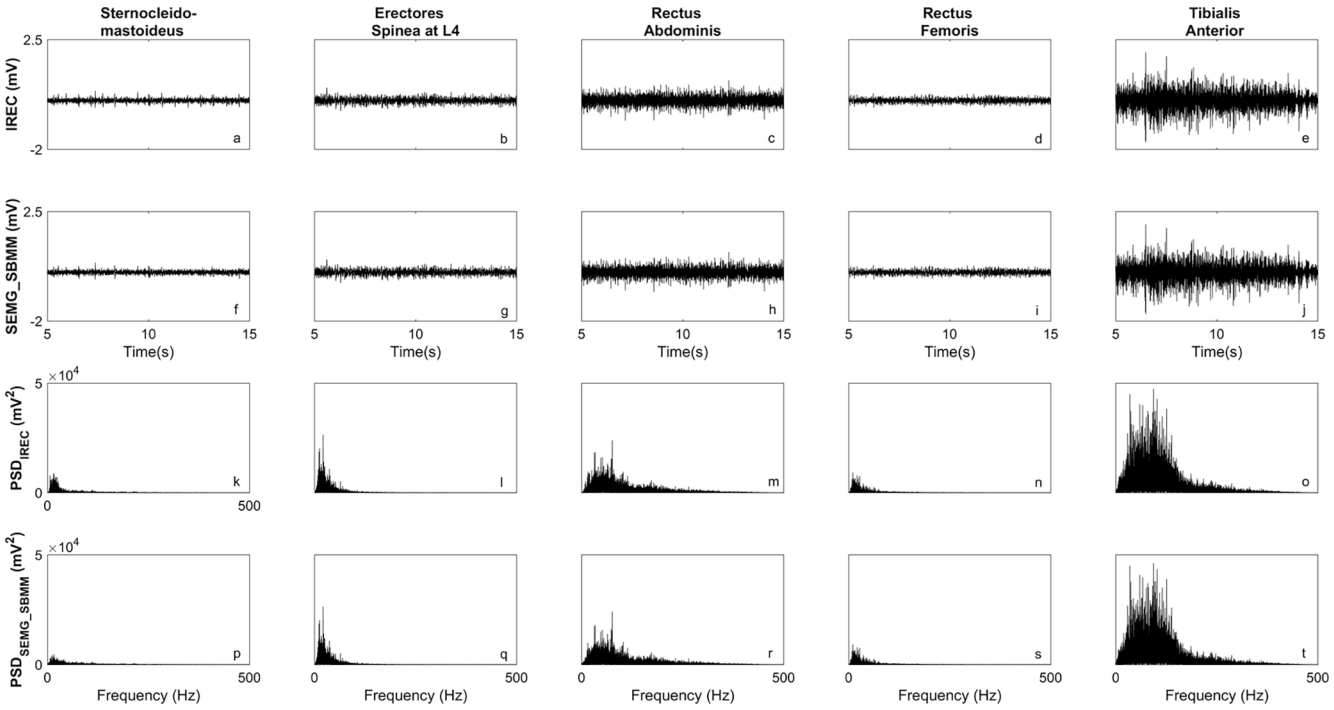

Fig. 4. Qualitative representation of signals involved in the electrophysiological study in both time domain (panels a to j) and frequency domain (panels k to t). Isometric recording (IREC) represents the original acquisition during an isometric muscular contraction, which was submitted to Segmented-Beat Modulation Method based filtering procedure to get a clean surface electromyogram estimation (SEMG_SBMM). Power Spectrum Density of IREC (PSD_{IREC}; panels k to o) and SEMG_SBMM ($PSDS_{EMG\_SBMM}$; panels p to t) were obtained by Fourier transform.

rectus femoris (Fig. 4d) and tibialis anterior (Fig. 4e), for both of which SNR = ∞. After application of the SBMM-based procedure to all IREC, obtained SEMG_SBMM (Fig. 4, panels f to j) signals were all characterized by SNR = ∞ but erectores spinae at L4 (SNR was 14[13; 17] dB), indicating a deletion of the ECG interference. Frequency spectra of these sample signals (Fig. 4, panels k to t) showed that, for f < 30 Hz, SEMG_SBMM spectra were lower (in case of sternocleidomastoideus, erectores spinae at L4 and rectus abdominis) or equal (in case of rectus femoris and tibialis anterior) to corresponding IREC spectra.

Quantitative results relative to all acquisitions of the electrophysiological study are reported in Table 2 and Table 3. Compared to IREC, SEMG_SBMM was typically characterized by a higher SNR, higher MdnF and lower PSD%(0,30), in accordance with the fact that application of SBMM removes ECG interference affecting IREC, with ECG interference having a frequency content mostly under 30 Hz. PSD %(30,450) trend over electrophysiological cases was complementary.

General evaluation of SEMG frequency content could be performed by analyzing results relative to SEMG_SBMM (Table 3). By considering all acquisitions in all muscles of all subjects, median MdnF and PSD %(0,30) were 74 Hz and 18% (last row and last column of Table 3), respectively, not significantly different from the corresponding values relative to single muscles (last row of each column of Table 3) or single subjects (last column of each row in Table 3). Both IREC and SEMG_SBMM had no fatigue effects, reflected in median FF values lower that 15% (7%).

Inter-muscle variability within a single subject (27[19; 30] Hz for MdnF and 15[10; 23] % for PSD%(0,30)) was comparable to inter-subject variability within a single muscle (35[19; 55] Hz for MdnF and 18[14; 31] % for PSD%(0,30)), as also depicted in Fig. 5. Both inter-muscle and inter-subject variabilities were significantly higher than intra-subject variability among different acquisitions (6[3; 14] Hz for MdnF and 4[1; 7] % for PSD%(0,30); $P < 10^{-3}$).

4. Discussion

The present paper evaluated SEMG frequency content in young healthy subjects. To this aim isometric contractions, both simulated and real ones, were considered to avoid motion artifacts. ECG interference was deleted by means of the SBMM-based procedure [22–24].

Our SBMM [23,24] was recently proposed in a preliminary study as

Table 2
Quantitative characterization of IREC relative to subjects involved in the electrophysiological study. Variable distributions were described in terms of median [25th percentiles; 75th percentiles] computed over acquisitions (subject 1 to 10), over subjects or over muscles. PSD%(30,450) not reported because, by definition, equal to 100-PSD%(0,30).

Subject	Variable	Sternocleidomastoideus	Erectores spinae at L4	Rectus abdominis	Rectus femoris	Tibialis anterior	All Muscles
1	SNR (dB)	9[8; 10]	11[11; 12]	8[8; 9]	∞[∞; ∞]	13[13; 13]	12[10; ∞]
	MdnF (Hz)	99[84; 105]	68[67; 68]	30[29; 36]	36[29; 37]	76[55; 79]	66[32; 75]
	FF(%)	11	10	12	11	1	11[10; 11]
	PSD%(0,30)	9[9; 12]	10[9; 11]	50[44; 52]	44[43; 54]	20[20; 32]	20[10; 44]
2	SNR (dB)	16[12; 17]	10[8; 10]	2[1; 2]	∞[∞; ∞]	∞[∞; ∞]	16[7; ∞]
	MdnF (Hz)	131[120; 138]	92[89; 101]	100[97; 111]	57[55; 58]	68[68; 70]	92[67; 109]
	FF(%)	7	6	7	8	6	7[6; 7]
	PSD%(0,30)	3[2; 5]	6[6; 7]	24[22; 25]	27[23; 29]	13[13; 19]	13[6; 24]
3	SNR (dB)	−4[-5;-3]	11[11; 11]	8[8; 8]	13[13; 13]	12[12; 12]	11[8; ∞]
	MdnF (Hz)	26[26; 29]	29[28; 29]	78[67; 80]	29[28; 31]	82[61; 83]	31[28; 72]
	FF(%)	6	13	9	9	2	9[6; 9]
	PSD%(0,30)	55[52; 56]	52[51; 54]	14[13; 21]	52[49; 54]	15[14; 28]	49[18; 54]
4	SNR (dB)	3[3; 3]	12[11; 12]	8[7; 9]	∞[∞; ∞]	13[13; 13]	12[7; ∞]
	MdnF (Hz)	68[64; 68]	87[85; 87]	42[41; 42]	84[83; 84]	84[78; 85]	82[63; 84]
	FF(%)	3	3	1	1	1	1[1; 3]
	PSD%(0,30)	22[21; 22]	6[6; 6]	40[40; 41]	11[11; 12]	11[11; 14]	12[10; 22]
5	SNR (dB)	3[3; 8]	8[8; 8]	3[3; 4]	∞[∞; ∞]	∞[∞; ∞]	8[3; ∞]
	MdnF (Hz)	60[37; 73]	80[75; 83]	64[59; 66]	67[63; 84]	92[92; 93]	70[61; 90]
	FF(%)	30	30	5	6	5	6[5; 30]
	PSD%(0,30)	24[19; 52]	9[8; 10]	28[28; 30]	16[14; 18]	9[9; 10]	15[10; 27]
6	SNR (dB)	1[0; 2]	9[8; 9]	−6[-6;-6]	∞[∞; ∞]	∞[∞; ∞]	9[0; ∞]
	MdnF (Hz)	25[24; 27]	68[66; 70]	26[26; 26]	52[52; 52]	108[94; 113]	52[26; 71]
	FF(%)	7	19	1	3	3	3[3; 7]
	PSD%(0,30)	56[53; 57]	12[11; 12]	55[55; 56]	31[31; 31]	13[12; 17]	31[13; 55]
7	SNR (dB)	5[3; 6]	12[12; 13]	5[5; 5]	∞[∞; ∞]	∞[∞; ∞]	12[5; ∞]
	MdnF (Hz)	75[69; 94]	79[79; 81]	76[66; 77]	51[51; 61]	69[67; 69]	70[64; 79]
	FF(%)	16	12	7	8	1	8[7; 12]
	PSD%(0,30)	19[15; 23]	7[7; 8]	24[24; 27]	35[30; 35]	28[27; 28]	25[13; 28]
8	SNR (dB)	0[-1; 1]	10[9; 11]	2[2; 2]	∞[∞; ∞]	∞[∞; ∞]	10[2; ∞]
	MdnF (Hz)	57[55; 94]	80[78; 82]	110[83; 110]	55[54; 59]	78[76; 80]	75[57; 83]
	FF(%)	35	26	12	13	3	13[12; 26]
	PSD%(0,30)	31[22; 31]	7[6; 7]	21[20; 26]	30[29; 33]	20[20; 21]	21[14; 31]
9	SNR (dB)	22[19; 25]	4[3; 5]	11[10; 11]	∞[∞; ∞]	14[13; 15]	15[10; ∞]
	MdnF (Hz)	12[12; 14]	16[15; 17]	65[65; 65]	85[84; 89]	104[103; 106]	65[15; 92]
	FF(%)	7	6	0	0	6	6[0; 6]
	PSD%(0,30)	86[79; 88]	76[72; 76]	10[9; 10]	18[16; 19]	6[6; 7]	18[9; 75]
10	SNR (dB)	11[11; 11]	−2[-3;-1]	8[7; 8]	∞[∞; ∞]	13[13; 14]	11[7; 14]
	MdnF (Hz)	90[87; 91]	29[29; 30]	72[65; 74]	82[80; 82]	93[91; 95]	82[62; 90]
	FF(%)	2	2	6	5	2	2[2; 5]
	PSD%(0,30)	14[13; 14]	52[50; 52]	17[14; 20]	8[8; 8]	6[5; 6]	13[8; 22]
All subjects	SNR (dB)	5[1; 11]	10[8; 12]	6[2; 9]	∞[∞; ∞]	∞[14; ∞]	12[6; ∞]
	MdnF (Hz)	66[26; 91]	71[30; 84]	65[42; 78]	58[51; 82]	82[72; 93]	69[51; 84]
	FF(%)	7[6; 15]	11[6; 18]	7[2; 9]	7[4; 9]	3[1; 5]	7[4; 9]
	PSD%(0,30)	22[13; 55]	9[7; 50]	27[20; 40]	28[14; 36]	13[9; 21]	20[10; 36]

FF: Fatigue Factor; MdnF: Median Frequency; PSD: Power Spectrum Density; SNR: Signal-to-Noise Ratio.

Table 3
Quantitative characterization of SEMG_SBMM relative to subjects involved in the electrophysiological study. Variable distributions were described in terms of median [25th percentiles; 75th percentiles] computed over acquisitions (subject 1 to 10), over subjects or over muscles. PSD%(30,450) not reported because, by definition, equal to 100-PSD%(0,30).

Subject	Variable	Sternocleidomastoideus	Erectores spinae at L4	Rectus abdominis	Rectus femoris	Tibialis anterior	All Muscles
1	SNR (dB)	∞[∞; ∞]	14[14; 15]	9[7; 11]	∞[∞; ∞]	∞[∞; ∞]	∞[15; ∞]
	MdnF (Hz)	100[86; 106]	68[67; 69]	31[30; 37]	36[29; 37]	76[55; 79]	67[35; 75]
	FF(%)	10	11	12	1	1	1[1; 11]
	PSD%(0,30)	9[9; 11]	10[9; 11]	49[43; 51]	44[43; 54]	20[19; 39]	20[10; 44]
2	SNR (dB)	18[17; 19]	14[14; 14]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[15; ∞]
	MdnF (Hz)	131[122; 139]	93[90; 103]	121[115; 134]	57[55; 58]	68[68; 70]	93[67; 119]
	FF(%)	6	8	7	6	7	7[6; 7]
	PSD%(0,30)	3[2; 4]	6[5; 7]	17[14; 18]	27[23; 29]	13[13; 19]	13[6; 20]
3	SNR (dB)	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]
	MdnF (Hz)	56[46; 60]	29[28; 29]	79[68; 82]	29[28; 31]	82[61; 83]	40[29; 76]
	FF(%)	13	9	9	2	2	9[2; 9]
	PSD%(0,30)	31[30; 36]	52[51; 53]	12[12; 20]	52[49; 53]	15[14; 28]	40[18; 52]
4	SNR (dB)	∞[∞; ∞]	13[12; 13]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[13; ∞]
	MdnF (Hz)	75[71; 76]	87[86; 87]	44[44; 45]	84[83; 84]	84[78; 86]	82[68; 84]
	FF(%)	3	1	1	1	1	1[1; 1]
	PSD%(0,30)	17[16; 17]	6[6; 6]	39[39; 40]	11[11; 12]	11[11; 14]	12[10; 18]
5	SNR (dB)	∞[∞; ∞]	12[12; 13]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]
	MdnF (Hz)	64[40; 78]	81[76; 85]	71[66; 74]	67[63; 84]	92[92; 93]	78[65; 92]
	FF(%)	30	6	5	5	5	5[5; 6]
	PSD%(0,30)	18[15; 49]	9[8; 10]	25[25; 27]	16[14; 18]	9[9; 10]	13[10; 23]
6	SNR (dB)	∞[∞; ∞]	15[13; 16]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]
	MdnF (Hz)	36[32; 45]	70[68; 72]	106[104; 110]	52[52; 52]	108[94; 113]	70[52; 105]
	FF(%)	19	3	1	3	3	3[3; 3]
	PSD%(0,30)	46[41; 49]	11[10; 11]	20[20; 21]	31[31; 31]	13[12; 17]	21[12; 31]
7	SNR (dB)	∞[∞; ∞]	14[14; 15]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]
	MdnF (Hz)	81[79; 99]	80[79; 82]	83[72; 85]	51[51; 61]	69[67; 69]	77[66; 83]
	FF(%)	12	8	7	1	1	7[1; 8]
	PSD%(0,30)	14[11; 15]	7[7; 8]	21[20; 24]	35[30; 35]	28[27; 28]	21[10; 28]
8	SNR (dB)	∞[∞; ∞]	14[14; 14]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[∞; ∞]
	MdnF (Hz)	74[68; 107]	82[78; 83]	125[95; 128]	55[54; 59]	78[76; 80]	75[63; 84]
	FF(%)	26	13	12	3	3	12[3; 13]
	PSD%(0,30)	20[13; 21]	6[6; 6]	16[15; 22]	30[29; 33]	20[20; 21]	20[9; 27]
9	SNR (dB)	31[27; 43]	∞[∞; ∞]	13[12; 13]	∞[∞; ∞]	∞[∞; ∞]	∞[25; ∞]
	MdnF (Hz)	12[12; 14]	16[16; 18]	66[66; 66]	85[84; 89]	104[103; 106]	66[15; 92]
	FF(%)	6	0	0	6	7	6[0; 6]
	PSD%(0,30)	85[79; 86]	74[68; 74]	9[9; 10]	18[16; 19]	6[6; 7]	18[9; 74]
10	SNR (dB)	∞[∞; ∞]	∞[∞; ∞]	14[13; 14]	∞[∞; ∞]	15[15; 15]	15[14; ∞]
	MdnF (Hz)	91[89; 92]	35[34; 39]	74[68; 76]	82[80; 82]	93[92; 95]	82[65; 91]
	FF(%)	2	5	6	2	8	5[2; 6]
	PSD%(0,30)	13[12; 13]	44[41; 45]	15[12; 19]	8[8; 8]	6[6; 6]	11[8; 21]
All subjects	SNR (dB)	∞[31; ∞]	14[13; 17]	∞[13; ∞]	∞[∞; ∞]	∞[∞; ∞]	∞[15; ∞]
	MdnF (Hz)	75[54; 93]	73[35; 84]	73[61; 102]	58[51; 82]	82[72; 93]	74[54; 87]
	FF (%)	11[6; 18]	7[4; 9]	7[2; 9]	3[1; 5]	3[1; 7]	7[5; 9]
	PSD%(0,30)	17[11; 37]	9[7; 44]	21[14; 28]	28[14; 36]	13[9; 21]	18[10; 31]

FF: Fatigue Factor; MdnF: Median Frequency; PSD: Power Spectrum Density; SNR: Signal-to-Noise Ratio.

an efficient tool to evaluate low-frequency spectral content in SEMG [22]. SBMM is a template-based technique able to extract ECG even when completely covered by other signals of both physiological and not physiological origin. In SEMG applications, SBMM may be used for estimating ECG interference affecting a recording, interference that is then deleted by subtraction. A further advantage of using SBMM is that it allows to separate (and thus to keep for further scopes) superimposed ECG and SEMG signals (estimation errors < 6.0%) [34] in case of both signals being of interest. On the other hand, SBMM requires knowledge of timing of R-peak positions, which is not directly available from SEMG recordings. Thus, application of SBMM-based filtering procedures for ECG deletion from a SEMG recording requires a dedicated simultaneous recording of the heart activity, which adds complexity to standard SEMG tests.

The simulation study demonstrated that the SBMM-based procedure is able to accurately remove ECG interference independently by its amplitude, by maintaining the SEMG low-frequency content. IREC remains unaltered in case of absence of interference (SNR of SEMG_SBMM equal to ∞ in all cases; Table 1). In addition, filtering does not significantly alter SEMG spectrum. Indeed, SEMG_SBMM MdnF was always equal to SimSEMG MdnF (that is 120 Hz; Table 1). In addition, SEMG_SBMM and SimSEMG PSD% distributions in low- and main-frequency bands differed of 2% at most. Specifically, such difference was 2% only in correspondence of a low SNR (5 dB; SIM1) that indicates a very high amplitude ECG interference; it was 0.0% in the other two simulated cases (SIM2 and SIM3). Thus, ECG removal altered the spectral content of the simulated recording (which included simulated SEMG plus simulated ECG) and allowed recovering of the simulated SEMG spectral content without distortions.

The electrophysiological study involved 10 young healthy subjects. The number of involved subjects was kept low on purpose to have the possibility of showing results relative to each participant. Each subject was asked to maximally extend the dominant arm perpendicularly to the trunk for at least 30 s. Bipolar electrodes were used, but application of our SBMM-based procedure is independent from used electrode type.

The movement performed by the subjects of this study is analogous to that performed during a Functional-Reach test [25–27] and allows to get isometric contractions of both upper and lower body muscles, from head to limbs. Isometric contractions were considered in order to understand if there is a significant amount of spectral components in the

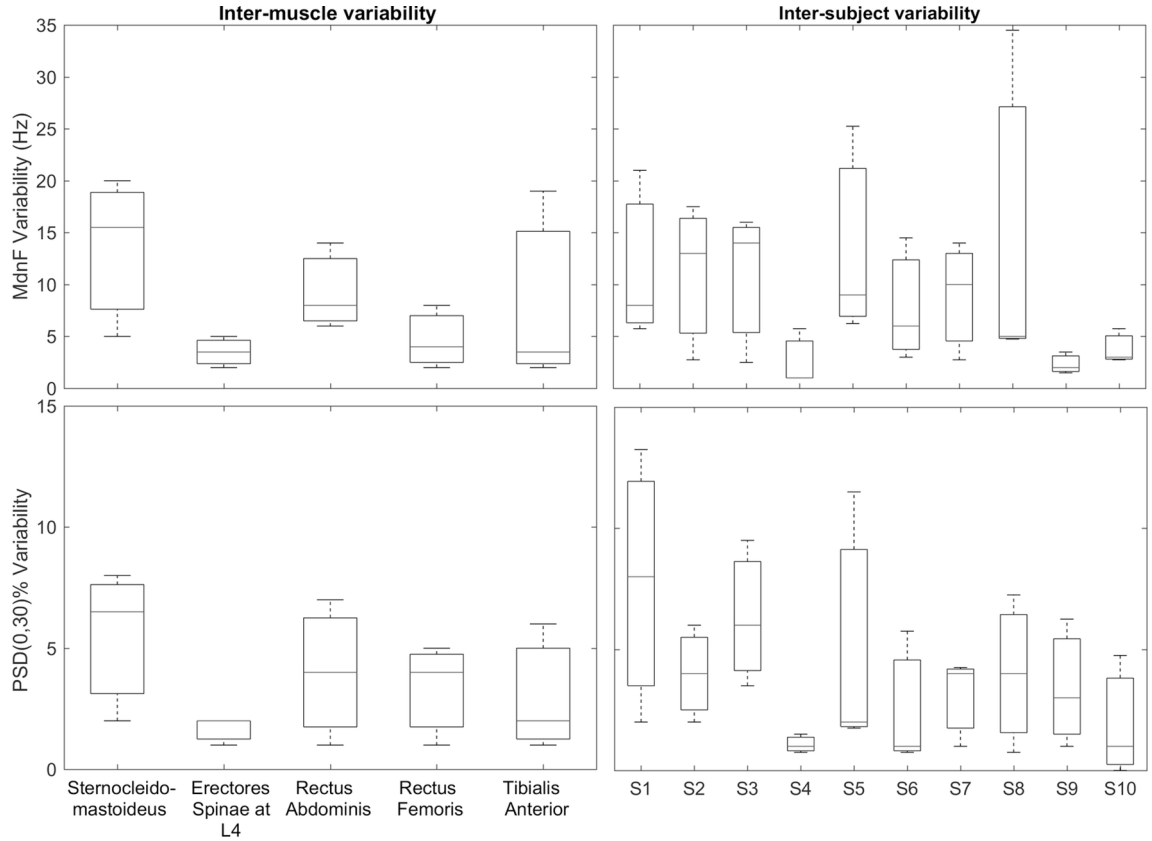

Fig. 5. Whisker diagrams (showing median, 25th and 75th percentiles, and range) relative MdnF and PSD%(0,30) variable, showing their inter-muscle variability and inter-subject variability (S1 to S10 indicated the 10 subjects involved in the electrophysiological study).

low-frequency band exclusively related to SEMG and not to ECG or motion artifact. Although here the SBMM-based procedure was applied to isometric SEMG recordings for ECG removal, it can also be applied to non-isometric SEMG recordings possibly affected by motion artifacts. In the latter case SBMM-based procedure will still delete ECG interference but will have no effect on motion artifacts; other specifically designed procedures should be designed to remove motion artifacts without distorting SEMG spectrum, as done by low-pass filtering.

In this study, SEMG recordings were acquired from isometric contractions of five muscles, which are sternocleidomastoideus, erectores spinae at L4, rectus abdominis, rectus femoris and tibialis anterior. Overall 50 (10 subjects by 5 muscles) muscles were analyzed. Since each acquisition was repeated three times, in total 150 measurements were performed. Results of the electrophysiological study confirmed the goodness of the SBMM-based procedure to remove ECG interference. Indeed, IREC relative to muscles close to the heart showed a lower SNR, which significantly increased after ECG-interference deletion. On the contrary, IREC relative to muscles far from the heart showed a higher SNR which remained unaltered after ECG-interference cancellation, confirming that such interference was negligible. More interestingly, results of the electrophysiological study provided some interesting insights on SEMG low-frequency spectral content. Median (over all subjects and muscles) MdnF was 74 Hz, in agreement with the 70 ÷ 100 Hz SEMG MdnF range proposed by De Luca [7,20]. Median PSD%(0,30) was 18%, indicating that almost a fifth of spectral content falls in the low-frequency band, and thus should not be neglected, especially by considering that this value represents an underestimation due to the integration of a 10 Hz high-pass filter in the Step32 system (which could not be turned off). Rather, specifically designed procedures should be considered to remove low-frequency interferences and motion artifacts from surface electromyographic recordings (analogously to what was done in digital electrocardiography), since such band could provide important physiological/clinical information, which is impossible to obtain at the present time due to use of low-pass filtering. The procedure proposed here uses the first version of SBMM [23,24], originally designed to extract ECG tracings characterized by sinus rhythm. Presence of atrial and ventricular premature beats would locally create artifacts in the SEMG_SBMM. Still, a second version of SBMM, including different template beats allowing reconstruction of sinus beats as well as premature atrial and ventricular beats is under development and will be published in the near future. Consequently, the SBMM-procedure proposed here to remove ECG interferences from SEMG recordings remains valid but use of the latest published SBMM version is recommended for an optimal performance.

If median MdnF and median PSD%(0,30) were not statistically different over muscles and subjects (Table 3), median inter-muscle variability (27 Hz) and median inter-subject variability (35 Hz) were relatively high and significantly higher than median intra-subject variability (6 Hz). Inter-muscle and inter-subject variabilities are possibly due to different muscle size and to a different level of activation during movement. This assumption will be further investigated in future studies, some of which will have also to be finalized to evaluate the possibility of defining baseline low-frequency content for each muscle of a subject; variations from baseline level could indeed be addressed to muscle fiber type, fatigue, training, pathologies and others [7,21].

5. Conclusions

In young healthy subjects, SEMG low-frequency spectral content is characterized by a median frequency of 74 Hz, whereas components below 30 Hz are 18%, on average. The latter cannot be addressed to ECG interference and/or motion artifacts, so that SEMG filtering procedures should avoid canceling or distorting the SEMG low-frequency band not to limit subsequent SEMG scope and applications finalized to

evaluate clinical content of this band. Future studies will investigate the importance of the low-frequency components in dynamic and clinical applications, especially but not exclusively in motor-unit analysis and in SEMG fatigue-effect comprehension.

Acknowledgement

Nothing to declare.

Appendix

A. Segmented-Beat Modulation Method

SBMM is an ECG filtering technique which requires knowledge of R-peak positions to be applied [23,24]. It is able to extract a clean ECG from a noisy recording, where the noise can be either external (such as power-line noise) or physiological (such as electromyographic noise). SBMM is a template-based method that initially performs template computation (Step 1) and then ECG reconstruction (Step 2) by template concatenation (Fig. A.1a). Differently from the other template-based methods, SBMM is able to keep track of the heart-rate variability thanks to a unique modulation/demodulation procedure applied to each single cardiac cycle (CC).

A.1. Step 1: template computation

To compute the template (Fig. A.1b, left panel) all beats in the noisy ECG are identified thank to the R-peak sequence, which is also used to compute the median RR interval (MRR). By relying on the experimental observation that, in first approximation, QRS complex has a constant duration whereas the duration of all other ECG waves are linearly dependent on the instantaneous heart rate (that is on previous RR interval) [35], SBMM divides each CC into two segments: 1) QRS segment, identified ± ΔT ms around the R peak, and thus of fixed length equal to 2·ΔT ms; and 2) TUP segment, identified within ΔT ms after the R peak and ΔT ms before of the subsequent R peak, and thus of variable length equal to the difference between CC and QRS durations. Successively, TUP segments undergo a modulation process (stretching or compression) which forces the belonging CC to have a length equal to MRR. Eventually, the template or median CC (MCC) is computed as the median of all modulated CC reconstructed using the original QRS segments and the modulated TUP segments. If MCC is really a representation of an ECG beat, MCC amplitude (maximum minus minimum) of the QRS complex has to be higher than 10% of four times MCC standard deviation. This condition is typically not satisfied when no ECG interference affects a SEMG recording or when ECG interference is so small to be negligible. In these cases, MCC is considered noise rather than a representation of an ECG beat. Consequently, MCC is set equal to zero.

A.2. Step 2: electrocardiogram reconstruction

The reconstruction of the clean ECG (Fig. A.1b, right panel) is performed by MCC N-fold concatenation (N is the number of CC in the noisy ECG) after median TUP (MTUP) segment demodulation (compression or stretching). Demodulation forces each reconstructed CC to have a length equal to that of the corresponding CC in the original noisy recording. Eventually, optimization processes based on cross-correlation maximization and distance minimization between reconstructed and original CC are performed to adjust to second-order inter-beat variations of CC waveforms.

Conflicts of interest

Authors disclose no financial and personal relationships with other people or organizations that could inappropriately influence (bias) their work.

Ethical statement

The present study was undertaken in compliance with the ethical principles of Helsinki Declaration and approved by the institutional expert committee; all participants gave their informed consent prior to testing.

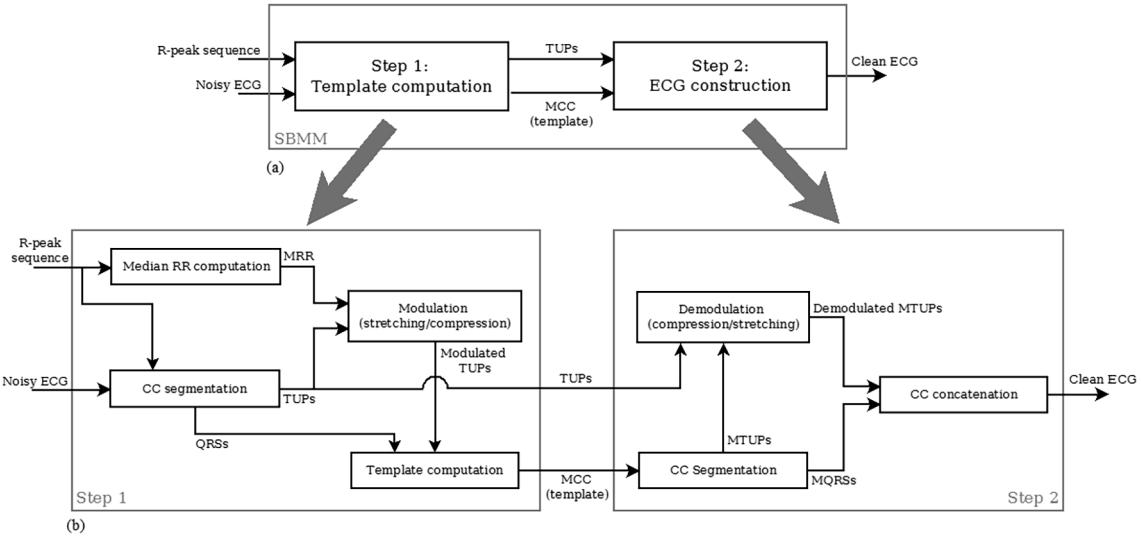

Fig. A.1. Block diagram of the Segmented-Beat Modulation Method (SBMM) filtering technique for getting a clean electrocardiogram (ECG) from a noisy recording.

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