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# Heel-strike in walking: assessment of potential sources of intra- and intersubject variability in the activation patterns of muscles stabilizing the knee joint

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#### 48 **1 Introduction**

Electromyography (EMG) is frequently used in studies of neuro-muscular motor control. The number of degrees of freedom in the musculoskeletal system allows for a wide variety of possible movements, and thus requires a complex control system. The flexibility of
the musculoskeletal system is illustrated by high step-to-step and inter-subject variability in muscle activation patterns (Araujo et al., 2000; Nair et al., 2010; Winter and Yack, 1987), which can be attributed to anatomical, neuro-muscular, and physiological reasons, among others (De Luca, 1997). However, it remains unknown how different sources of variability
contribute to the overall variability in the EMG signals. This study employed a principal component analysis (PCA) to assess the characteristics of EMG variability, and tested hypotheses derived from conceptual considerations of potential mechanical sources of EMG variability associated with the heel-strike event in walking.

Even though EMG signals are highly individual, temporal features of processed EMG signals, such as rhythmicity and timing of muscle activation, are common between individuals (Bizzi et al., 2008; Guidetti et al., 1996; Huber et al., 2011; Hug et al., 2010; Stirling et al., 2011). The application of PCA to EMG signals has enabled extraction of
information concerning neuro-muscular processes (Astephen Wilson et al., 2011; Ivanenko et al., 2004), the nature of the movement's coordination (Cappellini et al., 2006; Klarner et al., 2010; Sadeghi et al., 2000, von Tscharner, 2002; von Tscharner and Goepfert, 2003a), and mechanical efficiency (Blake and Wakeling, 2012; Blake et al., 2012). In the present study,
PCA was applied to EMG waveforms of the period from 200 ms before heel-strike to 200 ms after heel-strike, in level walking.

Within the analyzed movement, the heel-strike event is likely to be an important source of variability. Both foot placement angle (Murray et al., 1970) and the lateral component of the ground reaction force (Giaskas and Baltzpoulos, 1997) show high step-to-

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step variability at heel-strike. It appears that the neuro-muscular control system reacts and adjusts to the specific conditions at each heel-strike (Basmajian and De Luca, 1985) (i.e., mechanical sources of variability), and that these reactions are an important source of EMG
variability. The fastest mechanisms that could facilitate adjustments to the conditions at heel-strike are reflex circles, with a reaction time of 30-50 ms (Brooks, 1986; Schmidt and Lee, 1999; Williams et al., 2001). Therefore, a characteristic EMG feature indicating such reflex mechanisms would be maximum variability at approximately 30-50 ms after heel-strike
(Fig. 1a). Since reactions to the heel-strike event depend on the specific conditions of each walking step, they may be a source of inter-subject and intra-subject EMG variability. Muscle reactions require a low level of neuronal processing (Williams et al., 2001), and therefore, we hypothesize that these reactions are similar between subjects and that the shape of deviations

Another source of inter-subject variability could be the pre-activation of muscles before heel-strike (von Tscharner and Goepfert, 2003b). This pre-activation may be modulated by the so-called muscle tuning mechanism described by Nigg and Wakeling (Nigg, 2001; Nigg and Wakeling, 2001). They suggested that the impact at heel-strike could cause 88 potentially harmful shockwaves and vibrations within the body's soft tissues if these shockwaves could not be dampened at impact. However, they suggest that the neuro-muscular system "tunes" itself before impact such that the shockwaves are optimally dampened. 92 Therefore, a characteristic EMG feature associated with this mechanism would be a preactivation present at heel-strike that persists until reaction cycles can attune the muscles to the specific heel-strike event. As soon as the reaction cycles attenuate the system to the specific heel-strike conditions, one would expect a sharp decline in the activation (Fig. 1b). It has been suggested that muscle tuning is highly subject-specific because vibration and dampening 96 properties of the soft tissue packages depend on the mass and geometry (wobbling mass) of each individual's soft tissue compartments (Boyer and Nigg, 2006; Nigg and Liu, 1999; Pain and Challis, 2004, 2006). Therefore, the specific waveform shape associated with muscle tuning was not expected to correlate between subjects.

In summary, the primary aim of this study was to calculate and compare principal components (PCs) of intra-subject and inter-subject variability in knee muscle EMG waveforms at heel-strike. It was hypothesized that adaptation to the heel-strike event in walking is a major source of variability. Two waveform shapes with distinct characteristic features were proposed, based on conceptual considerations of how the neuro-muscular system might prepare for, or adapt to, the heel-strike event. A secondary aim was to determine if the calculated PCA waveforms showed the predicted characteristic features of the proposed waveforms.

2 Materials and Methods

## **2.1 Participants**

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The study group consisted of ten healthy female volunteers (age: 48 ± 7 years, body mass: 61.1 ± 4.9 kg, height: 1.64 ± 0.05 m [mean ± SD]) with no history of lower extremity surgery, no osteoarthritis of the hip, knee or ankle joints, and no neurological or musculoskeletal impairments. Volunteers were informed about the measurement procedure and provided written consent prior to participation. The local ethics committee approved the study.

## 2.2 Experimental design

The study protocol consisted of an instrumented three-dimensional gait analysis with 120 synchronous measurement of thigh muscle activity during level walking. A six-camera, 240 Hz motion capture system (Vicon MX13+, Oxford, UK) recorded the positions of retroreflective markers according to the Vicon Plug-In Gait model (kinematic model V 2.0, Vicon Motion Systems, Oxford, UK; Kadaba et al., 1990). The subjects walked barefoot at a comfortable, self-selected walking speed along a 10 m walkway ( $1.22 \pm 0.06$  m/s [mean  $\pm$  SD]).

## 2.3 EMG data recording

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Surface EMG signals of three quadriceps femoris muscles: rectus femoris (RF), vastus
medialis (VM), and vastus lateralis (VL); and two hamstring muscles: semitendinosus (ST) and long head of biceps femoris (BF) were recorded from the left thigh with bipolar Ag/AgCl surface electrodes (diameter: 10 mm, inter-electrode distance: 22 mm, Noraxon U.S.A Inc., Scottsdale, AZ, USA), in accordance with the SENIAM guidelines (Hermens et al., 2000).
The ground electrode was positioned over the tibial tuberosity. The electrodes were connected to single differential amplifiers (Biovision, Wehrheim, Germany. Bandwidth 10-700 Hz, gain range 1000-5000). Elastic net bandages (Elastofix, Typ B-25 m stretched, BSN medical

136 and electrodes in place. The EMG data were sampled at 2400 Hz without further processing.

## 2.4 Data processing

A time-frequency analysis, consisting of 13 non-linearly scaled wavelets (von Tscharner, 2000), yielded time and frequency distributions of the power of the EMG signal.

GmbH & Co. KG, Hamburg, Germany) were pulled over the thigh to keep cables, amplifiers,

140 The EMG power at each time frame was defined as the sum of the powers extracted from the wavelets with centre frequencies from 19 to 395 Hz. Wavelets with center frequencies lower than 19 Hz or higher than 395 Hz were omitted from the analysis, as they are known to be highly influenced by movement artifacts (Conforto et al., 1999) and high frequency noise 144 (e.g., 400 Hz power supplies), respectively. EMG power was analyzed from 200 ms before heel-strike to 200 ms after heel-strike, henceforth referred to as a waveform. Each waveform

was represented by 960 data points (sample frequency of 2400 Hz). Time 0 within the waveform was defined to be at heel-strike, which was determined from the vertical position of

- the heel marker. All waveforms were normalized by the integrated power; thus, the integrated power of the normalized waveforms was always 1. Due to this normalization, information about the degree of muscle activity is lost. Thus, the waveform shapes can be compared between subjects, but the amplitudes cannot. For each subject, 18 waveforms from the left leg were extracted, yielding a total of 180 waveforms available for further processing. Individual
- mean waveforms were calculated for each muscle by averaging the 18 waveforms of each subject. Group mean waveforms were computed for each muscle by averaging the 10 individual mean waveforms.
- The waveforms were stored in an *N* x *p* matrix, where *N* and *p* represent the number of waveforms and the number of data points in the waveforms, respectively. This matrix was denoted the input matrix (i.e., input matrix to the PCA), and the *N* waveforms were treated as vectors of a *p*-dimensional vector space. Two PCA procedures (Jolliffe, 2002), with differing
  types of input matrices, were performed. In both PCA calculations, the mean waveform of the input matrix was subtracted prior to calculation of the covariance matrix. In the "intra-subject PCA", the individual waveforms of each muscle and subject were assembled into an input matrix *M*<sub>18x960</sub>. A total of 50 intra-subject PCAs one for each subject and muscle were
  used to identify correlated deviations from the individual mean activation waveform of a given muscle (intra-subject variability). In addition, an "inter-subject PCA" was performed, by forming an input matrix *M*<sub>180x960</sub> from all 180 waveforms of a given muscle (10 subjects x 18 waveforms).
- The PCA yielded: (i) the eigenvectors of the covariance matrix of the input matrix, known as principal component vectors; (ii) the eigenvalues; and (iii) the loading factors, known as PC-scores (also known as PC-coefficients or weight factors). The PC-vectors represented correlated deviations from the mean waveform; that is, depending on which PCA

- 172 procedure was applied, they represented deviations from the individual mean waveform (intra-subject PCA) or from the group mean waveform (inter-subject PCA). The eigenvalues quantified the amount of variance explained by the corresponding PC. In this study, the eigenvalues were normalized by expressing them as a percentage of the sum of all
- 176 eigenvalues, i.e., as a percentage of the entire variability in the input matrix. The PC-scores were obtained by projecting each waveform onto the PC-vectors. PC-scores are a measure of how similar the measured waveforms are to a specific PC. The group mean waveforms, together with the lower-order PCs, captured the main features and modulations that are 180 common within the analyzed group of waveforms. The higher-order PCs represented fluctuations that were small in amplitude and/or not representative of the whole group.

As a similarity criterion, absolute values of the Pearson's correlation coefficient *r* were calculated for the 10 sets of intra-subject PC-vectors paired with the inter-subject PC-vectors for each of the five muscles.

All analyses were performed in Matlab (The MathWorks, Version R2011a, Natick, MA, USA) using custom-written programs.

## 188 **3 Results**

A visual representation of the inter-subject and intra-subject variability is shown in Fig. 2. The normalized eigenvalues of the first seven PCs for the inter-subject (Fig. 3, left) and intra-subject (Fig. 3, right) analyses identify two important results. Firstly, a substantial fraction of the EMG waveform variability was represented by only a few PCs. Across all five muscles, the first three inter-subject PCs accounted for 51.8 to 67.9% of the waveform variance, and the first three intra-subject PCs accounted for  $63.6 \pm 3.9\%$  to  $77.7 \pm 3.7\%$ (mean  $\pm$  SD across the five muscles) of the waveform variance. Thus, the individual

- 196 waveforms reconstructed from the group mean waveform and the first three inter-subject PC-vectors weighted by the mean individual PC-scores (mean over the 18 PC-scores of a subject)
  (Fig. 2, dashed line) contain enough information to mirror the basic activation pattern of an individual mean waveform (Fig. 2).
- The second result was that the normalized eigenvalues of the intra-subject analysis 200 agreed with those of the inter-subject analysis: PC<sub>2</sub>-eigenvalue range: 17.1-22.9% vs.  $19.2 \pm 2.0\%$  to  $23.7 \pm 4.8\%$  (all comparisons: inter vs. intra [mean  $\pm$  SD] across the five muscles), PC<sub>3</sub>: 9.8-12.8% vs.  $12.3 \pm 2.8\%$  to  $14.4 \pm 2.6\%$ , PC<sub>4</sub>: 8.2-9.5% vs.  $7.7 \pm 2.0\%$  to 204  $9.9 \pm 2.2\%$ , PC<sub>5</sub>: 5.0-7.3% vs.  $5.0 \pm 2.0\%$  to  $6.5 \pm 2.7\%$  PC<sub>6</sub>: 4.3-6.2% vs.  $3.5 \pm 1.5\%$  to  $4.3 \pm 1.6\%$ . The inter-subject analysis assessed the variability over 180 waveforms – with 10 times more potential sources of variability than the intra-subject analysis. The normalized eigenvalues of the inter-subject analysis might intuitively cover substantially less variability 208 than in the intra-subject analysis. However, a notable increase of the normalized eigenvalues was only displayed in PC<sub>1</sub>, for the muscles RF (from 22.2% vs.  $32.9 \pm 5.6\%$  [all comparisons: inter vs. intra [mean  $\pm$  SD] across the five muscles]), VM (28.9% vs. 39.0  $\pm$  6.7%), VL  $(30.2\% \text{ vs. } 37.9 \pm 7.1\%)$ , and BF  $(26.1\% \text{ vs. } 32.2 \pm 4.3\%)$ . The muscle ST showed similar 212 inter-subject and intra-subject variability in PC<sub>1</sub> (39.9% vs.  $36.0 \pm 5.0\%$ ). With regard to PCvector waveforms, PC<sub>1</sub>, displayed a strong correlation between intra-subject and inter-subject analyses, while higher-order PCs displayed moderate correlations (particularly PC<sub>2</sub>) or weak/no correlation (PC<sub>3</sub> and higher) (Fig. 4).
- Based on conceptual considerations, shapes with specific features were predicted (see Introduction) for the correlated deviations from the mean waveform, i.e. the PC-vectors. Such features were found in the first (Fig. 5) and second (Fig. 6) PC-vector. Figure 5 shows for the five analyzed muscles the shape of the inter-subject PC<sub>1</sub>-vector (top row) and gives a visual
  impression of how the waveforms changed when adding the inter-subject PC<sub>1</sub>-vectors

weighted with the individual mean PC<sub>1</sub>-scores of each subject to the group mean waveform (second row containing 10 graphs). Figure 6 shows the same for PC<sub>2</sub>. The main waveform feature predicted for variations in the reaction to heel-strike, an activation peak 30-50 ms after
heel-strike, was observed in PC<sub>1</sub> of the muscles VM, VL, ST, and BF (Fig. 5), and in PC<sub>2</sub> of the muscle RF (Fig. 6). These peaks occurred 51.3 ms, 40.0 ms, 52.5 ms, 42.5 ms, and 41.3 ms after heel-strike for the muscles RF, VM, VL, ST, and BF, respectively.

- The waveform features that were predicted for variations in the activation prior to 228 heel-strike – high muscle activation after heel-strike followed by a sharp decline at the same time where reactive mechanism peaked – were observed in PC<sub>1</sub> of the muscle RF (Fig. 5) and in PC<sub>2</sub> of the muscles VM, VL, ST, and BF (Fig. 6). All five muscles also showed a gradual increase in muscle activity in these PC-vectors, starting at least 50 ms before heel-strike for 232 the muscles RF, VM, VL and 80-100 ms before heel-strike for the muscles BF and ST. In the three knee extensor muscles (RF, VM, and VL), activation continued to increase after heelstrike, whereas for the knee flexors BF and ST, activation displayed a gradual decline after
- The up- or down-regulation of the changes in the waveform as characterized by PC<sub>1</sub> or PC<sub>2</sub> appeared to shift the peak in muscle activation from pre- to post-heel-strike (Figs. 5, 6). The higher-order inter-subject PC-vectors (PC<sub>3</sub> to PC<sub>7</sub>) showed multimodal shapes in all muscles with inter-peak time intervals in the range:  $53.3 \pm 7.8$  to  $120.8 \pm 11.6$  ms (Fig. 7).

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## **4** Discussion

heel-strike.

This study aimed to assess intra-subject and inter-subject variability of knee muscle EMG signals at heel-strike. The main findings were (i) a large fraction of the variability

- 244 (> 60%) was represented using only a few (three) PCs; (ii) the eigenvalues of the secondorder and higher-order PCs did not differ between the inter-subject and intra-subject analyses; and (iii) the shapes of the first two PC-vectors agreed well with predicted shapes derived from conceptual considerations.
- These findings suggest that the structure within a muscle activation pattern is not randomly organized. A significant fraction of the variability can be explained by a linear combination of distinct activation patterns. The PCA resolved the complex variability in the EMG signal (Fig. 2) into a small number of characteristic waveform deviation patterns (represented by PC-vectors). We hypothesized that these PC-vectors are not mere mathematical constructs, rather that they may be indicative of specific sources of variability affecting the EMG waveform. Specifically, the heel-strike event was identified as a major source of EMG variability, and it was found that for all five analyzed muscles, the shape of the first two PC-vectors exhibited features that had been predicted based on considerations of physiological pre-activation and reaction mechanisms.

Our results suggest that an up- or down-regulation of a reactive mechanism adjusting for the specific conditions present at heel-strike may be an important source of EMG variability represented in the PC<sub>1</sub>-vector of the muscles VM, VL, ST, and BF, and in the PC<sub>2</sub>vector of the muscle RF. The strong correlation between inter-subject and intra-subject PCvectors supports this interpretation. The delay between the heel-strike event and the peak in the PC-vector of the EMG activation was of the same order of magnitude as that previously reported in unconstrained overground walking (af Klint et al., 2010).

Some features associated with pre-activation in preparation for the heel-strike seemed to agree with the shape of the  $PC_1$ -vector calculated for the muscle RF and the  $PC_2$ -vectors obtained for VM, VL, BF, and ST. Pre-activation ensures that the knee has sufficient stiffness

- at the moment of heel-strike so as not to collapse; however, in all five muscles, the increase in activation level was only gradual and set in only 50 to 100 ms before heel-strike. Considering that the electromechanical delay between electrical stimulation and force production in a muscle is of the same order of magnitude (Cavanagh and Komi, 1979; Vos et al., 1990; Zhou
  et al., 1995), it seems questionable that the observed waveform would produce sufficient joint stiffness at the time of heel-strike. However, the observed PC-vector would fit to a mechanical model with variable knee stiffness: still relatively soft at the moment of heel-strike, then rapidly stiffening afterwards. Furthermore, variable knee stiffness due to gradually
  increasing muscle activation would lead to the soft tissue compartments around the knee vibrating with an increasing rather than a constant frequency. In fact, variable soft-tissue vibration frequencies were recently reported for running by Enders et al. (2012).
- Variability in the EMG signal is often interpreted as an indication of different neuronal control strategies (Ranganathan and Krishnan, 2012). However, if our interpretation is correct, a significant fraction of the EMG variability around the time of heel-strike can be attributed to processes that the neuro-muscular system uses to adjust for the mechanical conditions present. Moreover, our results suggest the possibility of distinguishing and separately investigating feedback and feed-forward mechanisms in the EMG signals of complex movements.

Some of the higher-order PC-vectors may be modulations of the two predicted waveforms as they also showed extreme values at approximately 30-50 ms after heel-strike. 288 Other waveforms exhibited a rhythmical variation with two or three oscillations. Recent studies have reported a heel-strike adjusted rhythm in the EMG signal at around 40 Hz in both running (Stirling et al., 2011) and walking (Huber et al., 2011). However, the exact pulse frequency differed between subjects, which might have led to an inter-subject EMG variability that contributed to the higher-order PC-vector waveforms.

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This study offers a new approach for analysis and interpretation of the inter-subject and intra-subject variability in the EMG signals of muscles that control the knee joint.Our interpretation is consistent with the present study's results; however, it is not necessarily the only possible interpretation. Further work is necessary to support or contradict these findings, such as an investigation of EMG variability when the heel-strike characteristics are systematically modified.

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## **Conflict of interest statement**

300 The authors report no potential conflict of interest associated with the study presented in this manuscript.

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#### **Figures**

- 424 Fig. 1: Predicted components of the EMG waveform derived from conceptual considerations of mechanisms that may play a role in the adaptation to the heel-strike. (a) Variability peak due to attenuation of muscle activity to the specific heel-strike conditions, based on an assumed reaction time of around 30 ms. (b) Pre-activation prior to impact, persisting until the system attenuates to the specific impact conditions. The predicted waveforms (solid lines) are affected by the normalization (in the present study, to unit power) that is necessary for
- comparison of EMG signals between subjects (De Luca, 1997; Frigo and Crenna, 2009). The dashed lines in (a) and (b) show a possibility of how the waveform might be distorted, taking
  into account that muscle activation in the analyzed muscles is mostly absent 200 ms before
- and 200 ms after heel-strike (Arsenault et al, 1986).
- Fig. 2. The individual mean waveforms for each of the ten subjects (s01 to s10, solid black
  lines), and the waveforms reconstructed from the group mean waveform and the first three inter-subject PC-vectors weighted by the individual mean PC-scores (dashed black lines) for the following muscles: rectus femoris, vastus medialis, vastus lateralis, semitendinosus, and biceps femoris. Intra-subject variability is indicated by gray shaded areas representing the
  standard error of the mean calculated for each subject. Each waveform has been scaled by its maximum range. Time 0 indicates heel-strike (vertical black line).

Fig. 3. The percentage variability explained by the first seven principal components (gray shaded bars) for the inter-subject PCA (left) and the intra-subject PCA (right, error bars

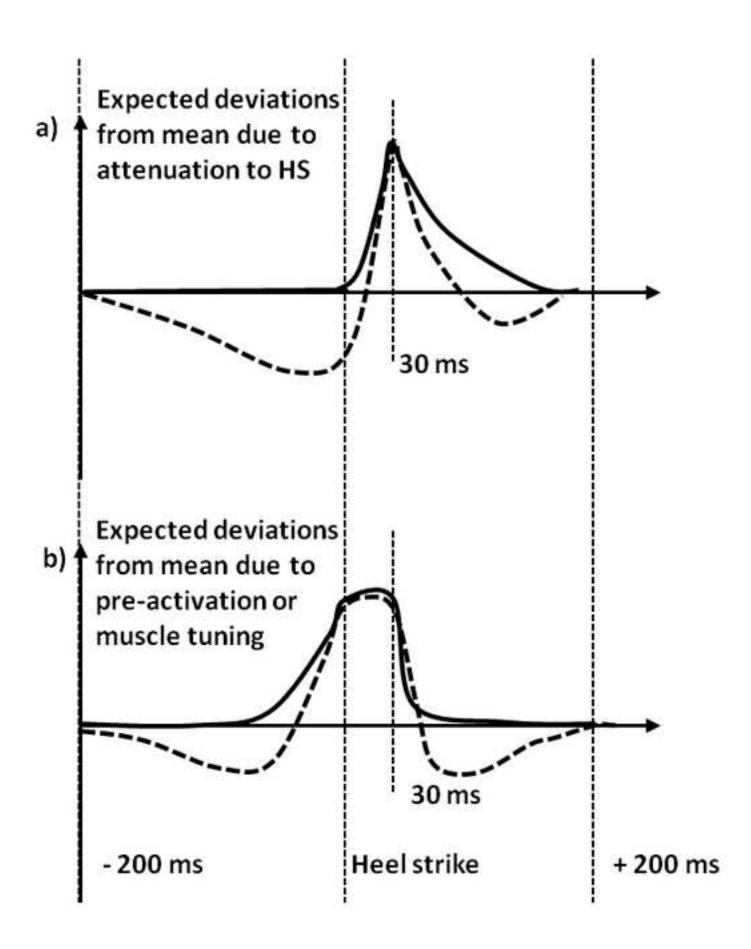
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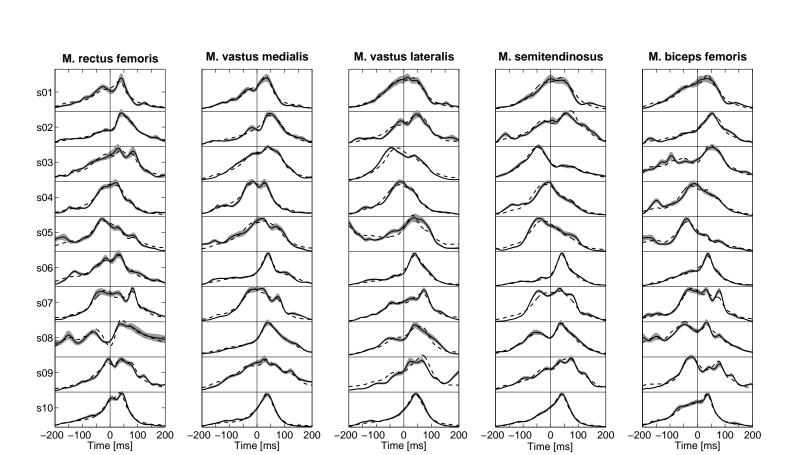
represent SDs across the 10 subjects) for the following muscles: rectus femoris (RF), vastus medialis (VM), vastus lateralis (VL), semitendinosus (ST), and biceps femoris (BF).

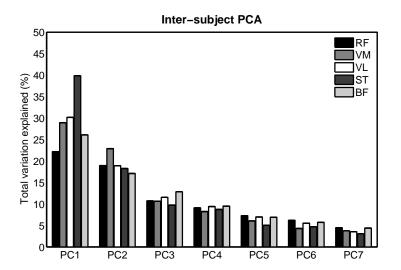
- 448 Fig. 4. Distributions of the correlation coefficients calculated for the 10 sets of intra-subject PC-vectors paired with the inter-subject PC-vector for the first seven PCs. Negative correlation coefficients have been inverted.
- 452 Fig. 5. Shape of the inter-subject PC<sub>1</sub>-vector (top row) and line graphs for each of the 10 subjects (s01 to s10) representing changes in the waveform when the PC<sub>1</sub>-vector (weighted by the individual mean PC<sub>1</sub>-score of each subject) was added to the group mean waveform (second row containing 10 graphs). The waveforms have been sorted from positive to 456 negative PC<sub>1</sub>-scores (displayed within each graph). Each waveform has been scaled by its maximum range.

Fig. 6. Shape of the inter-subject PC<sub>2</sub>-vector (top row) and line graphs for each of the
10 subjects (s01 to s10) representing changes in the waveform when the PC<sub>2</sub>-vector (weighted by the individual mean PC<sub>2</sub>-score of each subject) was added to the group mean waveform (second row containing 10 graphs). The waveforms have been sorted from positive to negative PC<sub>2</sub>-scores (displayed within each graph). Each line graph is scaled by its maximum
464 range.

Fig. 7. Higher-order inter-subject PC-vectors: PC<sub>3</sub>- (first row), PC<sub>4</sub>- (second row), PC<sub>5</sub>- (third row), PC<sub>6</sub>- (fourth row), and PC<sub>7</sub>-vectors (sixth row). Time 0 indicates heel-strike (vertical
black line).



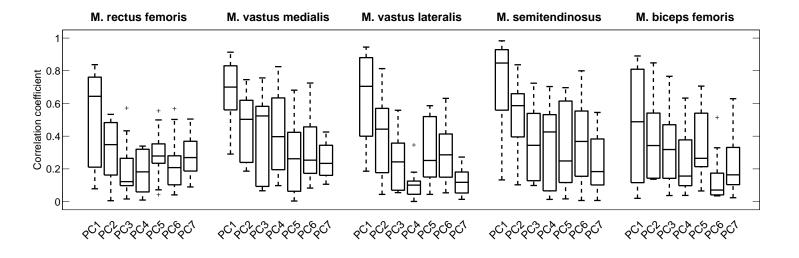




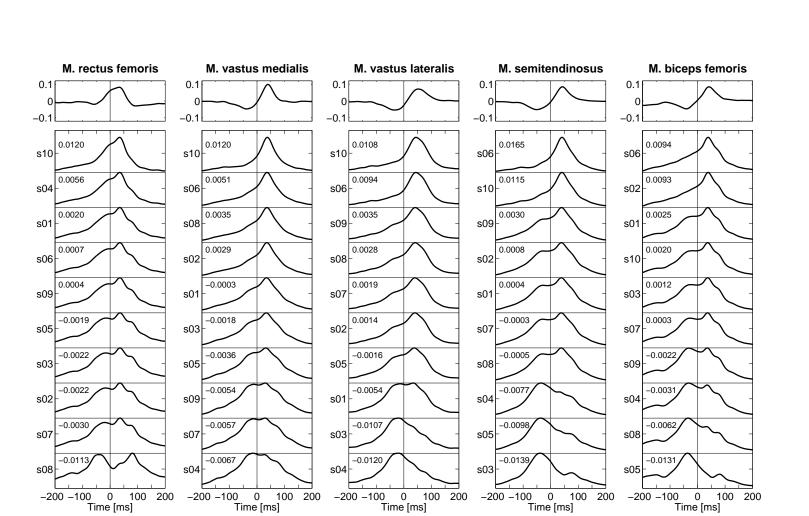
 $(S_{1}^{50} + I_{10}^{50}) = (S_{1}^{10} + I_{10}^{10}) = (S_{1}^{10} +$ 

Intra-subject PCA

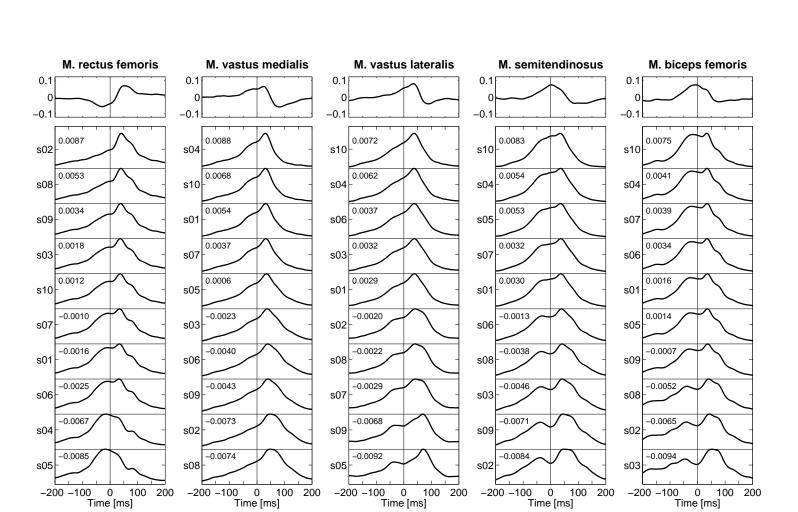












Fig#7

