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**Selectivity of conventional electrodes for recording motor evoked potentials: an investigation with high-density surface electromyography**

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**ABSTRACT:**

Introduction: The objective of this study was to determine whether motor evoked potentials (MEPs) elicited with transcranial magnetic stimulation and measured with conventional bipolar electromyography (EMG) are influenced by crosstalk from non-target muscles.

Methods: MEPs were recorded in healthy participants using conventional EMG electrodes placed over the extensor carpi radialis muscle (ECR) and high-density surface EMG (HDsEMG). Fifty MEPs at 120% resting and active motor threshold were recorded. To determine the contribution of ECR to the MEPs, the amplitude distribution across HDsEMG channels was correlated with EMG activity recorded during a wrist extension task.

Results: While the conventional EMG identified MEPs from ECR in >90% of the stimulations, HDsEMG revealed that spatial amplitude distribution representative of ECR activation was observed less frequently at rest than while holding a contraction ($P$<0.001).

Conclusions: MEPs recorded with conventional EMG may contain crosstalk from non-target muscles, especially when the stimulation is applied at rest.

**KEYWORDS:**

Transcranial Magnetic Stimulation; Motor evoked potential; Electromyography; High-density surface electromyography; Forearm; Wrist Extensor muscles.

## INTRODUCTION

Transcranial magnetic stimulation (TMS) is a useful way to non-invasively evaluate the excitability of the corticospinal system. Corticospinal excitability elicited by TMS is typically recorded with bipolar surface electromyography (EMG) placed over the peripheral muscle(s) of interest. While the focality of TMS is highly debated in the literature,[1,2] little attention has been paid to the selectivity of EMG recording systems, which are integral to the assessment of corticospinal excitability.

As surface EMG electrodes have a relatively large recording volume, it is possible that potentials measured from a pair of surface EMG electrodes may be influenced by muscles other than the one of interest.[3–5] This phenomenon, referred to as crosstalk,[6] could influence the amplitude of motor evoked potentials (MEP) elicited by TMS. Importantly, stimulation parameters such as localization of the hotspot in the primary motor cortex (M1), motor threshold, and stimulation intensity are routinely based on the amplitude of the MEP,[7–11] and hence they depend on whether the EMG recording reflects the activation of the target muscle. As the representation of individual muscles may overlap extensively in M1,[12] non-target muscles can also be stimulated. This highlights the importance of selective EMG recordings in TMS protocols, especially when electrodes are placed on the forearm where muscles with different motor functions are spatially adjacent.

High-density surface electromyography (HDsEMG) is a technique that utilizes several small electrodes placed closely over 1 or more muscles. When applied to the forearm, HDsEMG can be used to distinguish EMG amplitude distributions associated with activation of individual wrist extensor muscles.[13] This enables differentiation of MEPs from the extensor carpi radialis muscle (ECR) from those of the extensor digitorum communis muscle (EDC).[14] While this sophisticated analysis can be used to discriminate the activation of different muscles, HDsEMG is not commonly used in TMS studies, and a recent case study showed that MEPs from different forearm muscles could not be recorded selectively

using conventional electrodes.[3] These data illustrate that more information that characterizes the selectivity of conventional EMG would be of interest.

Thus, the aim of this study was to investigate whether: 1) MEPs observed with conventional bipolar surface EMG, hereafter referred to as conventional EMG, are representative of the activation of the target muscle, and 2) HDsEMG can help differentiate MEPs from the target muscle from those of surrounding muscles. We hypothesized that MEPs recorded with conventional EMG would contain crosstalk from muscles distant to the recording electrodes, whereas HDsEMG could differentiate MEPs from ECR from that of surrounding muscles.

**EXPERIMENTAL PROCEDURE**

Participants

Ten healthy right-handed individuals (5 women; 28 ± 4 years old) participated in this study. None of the participants had contraindications to TMS. All participants gave informed, written consent for the study. The study conformed to the standards set by the latest revision of the Declaration of Helsinki and was approved by the University of British Columbia Clinical Research Ethics Board.

Placement of the EMG electrodes

Participants sat comfortably in a chair during the testing session. The right ECR was localized through palpation during active wrist extensions. Optimal positioning of the conventional EMG system over ECR was confirmed using ultrasound (LogicScan 64 LT-1T, Telemed, Vilnius, Lithuania); in 3 subjects, the location chosen with palpation was near the edge of the EDC, so the position of the conventional electrodes was shifted medially approximately 10 mm to ensure the electrodes were within the ultrasound-guided boundaries of ECR. The HDsEMG grid comprised 5 individual arrays of 16 electrodes (ELSCH016, OT Bioelettronica, Torino, Italy; interelectrode distance: 10 mm) oriented along the forearm

(Figure 1) and kept in place using adhesive foam. The transverse inter-electrode distance was 20 mm; hence, a surface area of 150 mm (proximal to distal) by 80 mm (medial to lateral) was covered by electrodes. Conventional electrodes (H59P Cloth Electrodes, 7.8 $cm^2$; Covidien, Mansfield, MA, USA) were reduced to approximately 1 $cm^2$ to fit between the HDsEMG arrays and were inserted into foam with an inter-electrode distance of 30 mm (Figure 1) to ensure that both EMG recording systems could be performed simultaneously.

After standard skin preparation, the HDsEMG grid was applied to the skin so that the conventional EMG electrodes were placed over the ECR location, and the grid was oriented along the approximate direction of the ECR muscle fibers. In general, the grid spanned more than 70% of the length of the forearm and covered the following extensor muscles: ECR, EDC, and extensor carpi ulnaris (ECU). For the conventional EMG, the ground electrode was placed on the ulnar styloid. For the HDsEMG, the ground electrode was placed on the radial styloid; a second ground, needed for monopolar detection, was place on the olecranon. Conventional EMG data were collected with a 450 ms sweep from 100 ms before to 350 ms after TMS delivery using LabChart software (LabChart 7.0) and were sampled at 2k Hz, pre-amplified (1000x), and band-pass filtered at 10-1000 Hz using a Powerlab data acquisition system and a bioamplifer (AD instruments, Colorado Springs, CO). HDsEMG signals were collected in monopolar modality, amplified 200 times and digitized at 2048 samples/s using a 12 bit A/D converter (EMG-USB, OTBioelettronica, Torino, Italy). Pulses that identified the onset of the TMS stimulation were recorded by both systems simultaneously to ensure synchrony between both recording systems.

TMS and Neuronavigation

Single pulse TMS was delivered using a figure-of-eight shaped coil (Magstim 70 mm P/N 9790, Magstim Co., UK) connected to a Magstim $200^2$ stimulator (Magstim Co., UK). Each participant underwent a T1 anatomical magnetic resonance imaging (MRI) scan which was used for TMS targeting and position

monitoring using Brainsight™ neuronavigation (Rogue Research Inc., Montreal, QC, Canada), except for 1 individual (who had contraindications to MRI) where a standard anatomical brain was used. The 'hotspot' for eliciting MEPs in the contralateral ECR was found by localizing the cortical site in the hand/forearm M1 representation[15] where stimulation elicited the largest and most consistent MEPs as recorded with the conventional electrodes. Using conventional EMG, resting motor threshold was defined as the lowest stimulation intensity that elicited MEPs of at least 50 μV in 5 of 10 stimulations when TMS was applied at rest.[16] Active motor threshold was defined as the lowest intensity that elicited 5 of 10 responses of at least 200 μV when TMS was applied while holding a background ECR contraction;[10] using online EMG feedback, the contraction was maintained to produce EMG activity in ECR (average rectified value, ARV) close to 5% of that recorded during a maximal voluntary contraction. TMS pulses were delivered randomly at a rate between 0.15 and 0.2 Hz.

Data Collection Procedure

Fifty MEPs at rest and 50 MEPs while holding an isometric wrist extension were collected while single-pulse stimulation was applied at 120% of resting (RMT) and active (AMT) motor threshold, respectively. When MEPs were elicited while holding a contraction, 60s of rest was given after the twenty-fifth stimulation to limit the effects of fatigue. Selectivity of the TMS response was investigated by comparing the EMG amplitude distribution of MEPs to that of a selective activation of ECR. To selectively activate the ECR, participants performed a low-force, isometric wrist extension guided by visual feedback from the HDsEMG grid that was presented to participants as a colormap on a monitor. Once EMG activity in a single region previously identified as representative of selective ECR activation[13] was obtained, 5 seconds of EMG were collected.

Data processing:

Custom MATLAB scripts (Mathworks, Natick, MA) were used to identify the peak-to-peak amplitude of the MEPs for each participant. Conventional EMG and HDsEMG monopolar signals were filtered offline (fourth order Butterworth filter, 10-400 Hz). The amplitude distribution during the voluntary ECR activation was obtained by calculating the ARV for each channel of the grid, resulting in a matrix of 16x5 values, 1 value per HDsEMG electrode. For each stimulus, the MEP amplitude from the conventional EMG was measured as the peak-to-peak value of the response observed with a latency of approximately 18 ms. The MEP amplitude from HDsEMG grid was measured as the peak-to-peak value of the response in each channel. Examples are illustrated in supplementary figure S1, available online. For each participant, the number of MEPs elicited by the 50 stimuli with amplitude at least 50 µV at rest or larger than 200 µV with a background contraction was calculated.

Analysis 1: Concurrent validity between conventional EMG and HDsEMG

Three analyses were performed. First, we examined the concurrent validity between the 2 EMG systems. As it is known that both modality detection (e.g. monopolar vs. bipolar) and physical characteristics of the electrode (e.g. size) influence EMG amplitude measurements[17], a virtual bipolar recording was created from the HDsEMG to allow for statistical comparison between the 2 methods. Two virtual electrodes were created by averaging monopolar EMG signals from 2 groups of 4 channels around each of the conventional electrodes (Figure 1). A virtual bipolar detection was calculated as the difference between these 2 virtual electrodes. This resulted in a virtual bipolar detection collected with electrodes with physical characteristics (position, size, and distance between the electrodes) comparable to the conventional EMG used in this study. Similar to the conventional EMG, the MEP amplitude was calculated as the peak-to-peak value of the response. The average MEP amplitude of the 50 stimuli applied at rest and with a background contraction was compared between the conventional EMG and the virtual bipolar detection systems.

Analysis 2: Localization of MEPs to ECR (HDsEMG)

Second, the MEP amplitude distribution within the surface area of the HDsEMG was calculated. For each of the 50 stimuli at rest and with the background contraction, the peak-to-peak value of the response from each monopolar channel was averaged across stimuli, resulting in 2 matrices of 16x5 amplitude values representing the average EMG amplitude distribution across the forearm for MEPs at rest ($MEP_R$) and while holding a background contraction ($MEP_A$). Examples of these average MEPs can be observed in the top panels of supplementary figure S1. To identify the location of the ECR in the HDsEMG grid, the EMG amplitude (ARV) was calculated for each channel of the grid during the selective wrist extension. To measure whether the EMG activation elicited with TMS is localized in the ECR, the MEP amplitude distribution was correlated with EMG amplitude distribution observed during voluntary ECR activation (bidimensional correlation, MATLAB function; see statistical analyses below).

Analysis 3: Localization of MEPs to ECR (innervation zone)

Third, the presence of an innervation zone under the electrodes was used as a further analysis to confirm the identification of crosstalk from activation of ECR. When placed along a muscle, HDsEMG allows detection of the main innervation zone.[17,18] In muscles with fibers roughly parallel to the skin, this region can be identified visually as a phase reversal of the propagating action potential in single differential EMG signals.[18] Following a motor neuron discharge, depolarization of the muscle fiber starts from the innervation zone and propagates toward the tendons; for this reason, the innervation zone can be observed in the HDsEMG signals when the muscle under the electrodes is activated. If the recording was mostly crosstalk (rather than propagating potentials from the ECR), then no innervation zones would be observed. Virtual bipolar detections along the ECR were calculated from groups of monopolar electrodes in the same configuration used in Analysis 1, but located 1, 2, and 3 electrodes proximal or distal to the original location. This resulted in 7 virtual bipolar channels spaced 10 mm apart oriented along the ECR.

For voluntary ECR activation, $MEP_R$ and $MEP_A$ (signals averaged over the 50 stimuli), the HDsEMG was inspected for presence of an innervation zone; the channel where the innervation zone was identified was noted.

Statistical analysis:

For Analysis 1, the concurrent validity of conventional EMG and virtual bipolar detections was established using Intraclass Correlation Coefficient (ICC, relative agreement) and normalized Standard Error of Measurement (SEM, absolute agreement). For each participant for Analysis 2, a correlation coefficient was calculated to compare the EMG amplitude distributions observed in voluntary ECR activation with that of the MEP amplitude distribution for $MEP_R$ and $MEP_A$. Similar spatial distributions would result in a correlation coefficient (*R*) close to 1, while progressively smaller values represent larger differences between the 2 distributions (Figure 2). To determine whether either $MEP_R$ or $MEP_A$ was more selective for ECR, a paired *t*-test was run on the R values (after Z-score transformation). For Analysis 3, we reported in which channel the innervation zone was localized in ECR, $MEP_A$, and $MEP_R$. Statistical significance was set at $P < 0.05$.

**RESULTS**

Motor evoked potentials:

On average, RMT and AMT were 40 ± 12% and 32 ± 9% of the maximal stimulator output, respectively. Stereotaxic imaging confirmed that the TMS coil was held over the determined ECR 'hotspot' accurately throughout the experiment (average error: 0.28 ± 0.07 mm) with small variations of coil orientation (0.65 ± 0.32 degrees). Using conventional EMG, MEPs were observed in 93% of the stimulations at rest (549 ± 404 μV) and in 99% of the stimulations applied while holding a background contraction (1317 ± 1246 μV). As the amplitude of the monopolar EMG signal is much larger than that collected with bipolar electrodes (supplementary figure S1), the standard 50 μV and 200 μV thresholds could not be used to determine

whether or not an MEP was produced in the HDsEMG; visual observation confirmed that trials when the MEP did not reach the threshold using conventional EMG had little or no activity in the HDsEMG.

Analysis 1: Concurrent validity between conventional EMG and HDsEMG

The MEP amplitude observed in a virtual bipolar detection calculated from HDsEMG was comparable to that of the conventional EMG. Intraclass correlation coefficients (ICC) indicated that the relative agreement was better for $MEP_R$ [ICC (2, 50) = 0.99] than $MEP_A$ [ICC (2, 50) = 0.91], though the ICC of both testing conditions remained high. The absolute agreement was also better for $MEP_R$ (SEM% = 7.4%) than $MEP_A$ (SEM% = 26.1%). The relatively high SEM% of $MEP_A$ may have been the result of 1 participant having MEPs (~4700 μV) much larger than the others (range 500-1500 μV). Excluding this individual reduced the SEM% to 16.9% but lowered the ICC to 0.81; for both absolute and relative agreement the $MEP_A$ remained lower than $MEP_R$.

Analysis 2: Localization of MEPs to ECR (HDsEMG)

To investigate whether the response to TMS was predominantly localized in the ECR, the amplitude distribution over the forearm of the MEPs was correlated to that observed in a voluntary, selective ECR activation. Similar to the representative participant in Figure 2, the group data also showed that the amplitude distribution of MEPs while holding a background contraction ($MEP_A$) was correlated with the EMG distribution observed during selective ECR activation (R = 0.73 ± 0.13). Conversely, the correlation between $MEP_R$ and ECR activation was generally low (R = 0.23 ± 0.31). The correlation values between the amplitude distribution observed during selective ECR activation and $MEP_A$ were significantly larger than those between the ECR amplitude distribution and $MEP_R$ (Figure 3; mean difference: 0.51; paired *t*-test: $P < 0.001$).

Analysis 3: Localization of MEPs to ECR (innervation zone)

To further confirm identification of activation of ECR from crosstalk, we looked for the presence of an innervation zone in the HDsEMG. Phase opposition reversal of the action potentials in consecutive channels could be observed in 9 of 10 participants during voluntary wrist isometric extension contractions (Figure 4, Table 1). In the same participants, an innervation zone was observed in the MEPA recordings, typically in the same channel as the isometric wrist extension or in an adjacent channel; the phase opposition reversal could be identified clearly as the first peak was of different polarity in the channels proximal or distal to the innervation zone. The innervation zone during $MEP_R$ could be observed in only 1 participant (Fig. 4). In the majority of participants, channels showed a similar shape during $MEP_R$, with the first peak having the same polarity for all channels.

**DISCUSSION**

Our data suggest that MEPs observed with conventional EMG may not be specific for the muscle of interest in the forearm; this difference was observed with HDsEMG. While EMG responses observed at rest using conventional EMG were assumed to be MEPs from ECR, 2 separate analyses based on amplitude spatial distribution and presence of an innervation zone showed that a large amount of the EMG activity was instead generated by other muscles. Crosstalk was less prevalent when TMS was applied while holding a background contraction, likely due to facilitation of the ECR muscle. Implications for clinical studies are discussed below.

HDsEMG showed that muscles other than ECR were prevalently activated by TMS at rest; yet large MEPs, usually interpreted as ECR responses, were observed in the conventional EMG. As procedures for electrode placement, definition of the 'hotspot', and thresholds were based on MEPs collected with a pair of conventional EMG electrodes,[8–10,19] these results are generalizable to other studies that used TMS to elicit MEPs from ECR. Changes in MEP amplitude are difficult to interpret when there is crosstalk from other muscles. For instance, TMS is commonly used to assess corticospinal excitability in clinical

populations[7,8,19,20] and in association with interventions.[8,11] Thus it is possible that when the excitability of the target muscle is comparable between conditions, but other muscles are facilitated or inhibited, MEPs collected with conventional EMG may erroneously indicate that the corticospinal excitability of the target muscle was increased or decreased. Furthermore, if the excitability of the 2 muscles is changed in opposing directions, the MEP from the target muscle may misrepresent the effect of the intervention or between the populations tested. This is a finding that is particularly relevant in studies where compensation from other muscles may occur. This may be particularly problematic for the study of clinical populations, such as in the case of stroke, where it is known that neuromuscular control is abnormal.[20,21] Of note, the lack of selectivity of conventional EMG is likely not as relevant when ECR is facilitated with a background contraction or when the identification of MEPs from a specific wrist extensor is unnecessary. Future studies should investigate whether other parameters determined using TMS, such as the extent of muscle representation in the motor cortex, differs when estimated with HDsEMG or conventional EMG.

Past work, using a smaller grid and an analysis based on the centroid of the amplitude distribution, also found that ECR activity in $MEP_R$ was low and larger when the MEP response was facilitated with a background contraction.[14] We confirmed those results using a correlation analysis to determine the contribution of the ECR muscle to the distribution of MEP amplitude that enabled comparisons to be made in each participant. We also conducted a secondary analysis based on the ECR innervation zone. The similar location of the innervation zones during selective ECR contraction and in MEPs elicited with a background contraction confirmed that ECR contributed to the MEP. As no innervation zones could be identified in MEPs at rest, this secondary analysis further confirmed a low ECR contribution to MEPs evoked at rest.

The amplitude of the MEPs estimated with conventional EMG or virtual electrodes with similar size, position, and inter-electrode distance obtained from HDsEMG was equivalent (ICC > 0.9). As for the

absolute agreement, the SEM% was smaller at rest (7%) than when an active contraction was maintained (17-26%). In this study, the conventional and virtual bipolar electrodes were placed in a similar position, but it was not possible to physically place them in the same location. This may explain some of the differences in the EMG amplitude measured by the 2 detection systems. It is possible that these differences in EMG amplitude were observed more in $MEP_A$ than in $MEP_R$ because of different characteristics of the signal (i.e., propagating vs. non-propagating); this possibility requires further investigation.

This study also showed that MEPs from ECR can be differentiated from crosstalk by visual observation of a small number of HDsEMG channels. The location of the innervation zone in $MEP_A$ and voluntary wrist extension was comparable. Finding the innervation zone is simple, reliable, and requires no signal processing.[18] In addition, the use of small HDsEMG arrays (e.g. 8 contacts) is no more time-consuming than placement of a pair of electrodes. Thus visual observation of innervation zones using HDsEMG arrays could be integrated into TMS protocols.

**CONCLUSIONS**

Using HDsEMG, we show that MEPs observed in conventional bipolar EMG placed over the ECR may be generated in non-target muscles. This suggests that MEPs recorded with conventional bipolar electrodes are not always specific to the target forearm muscle, especially when MEPs are elicited at rest. Evoking an MEP during an active contraction may more likely reflect target muscle activation. Using conventional EMG, changes in MEPs recorded over ECR may partly reflect changes in corticospinal excitability of non-target muscles.

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**TABLE:**

Table 1: Location of the innervation zone.

| Participant | IZ ECR | IZ $MEP_R$ | IZ $MEP_A$ |
|---|---|---|---|
| **1** | 4 | - | 5 |
| **2** | 5 | - | 3 |
| **3** | 4 | - | 3 |
| **4** | 3 | - | 3 |
| **5** | 3 | - | 2 |
| **6** | 5 | - | 4 |
| **7** | 3 | - | 3 |
| **8** | 5 | - | 5 |
| **9** | - | - | - |
| **10** | 4 | 2 | 4 |

Identification of the innervation zone (IZ) in the voluntary contraction of ECR, in MEPs at rest ($MEP_R$) and while keeping a background contraction ($MEP_A$). Numbers represent the location of the channel where the innervation zone was located (1 is proximal); hyphens indicates that no innervation zone was observed.

**FIGURES:**

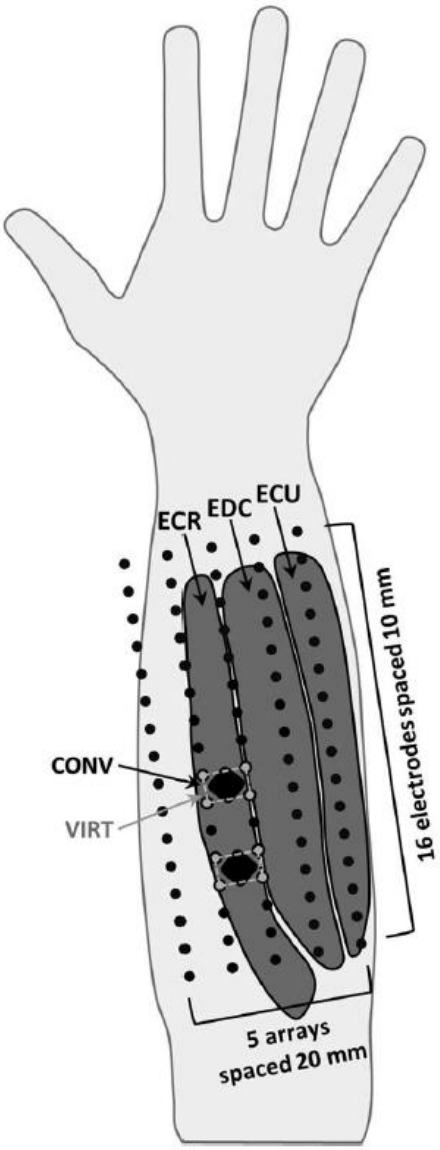


Figure 1: Experimental set-up. Dots identify HDsEMG electrodes; diamonds identify conventional bipolar electrodes. The HDsEMG channels used to create the virtual bipolar EMG are represented as grey dots surrounding the conventional electrodes; note how the size, position, and distance between virtual electrodes (VIRT, grey dashed lines) compares to that of the conventional bipolar electrodes (CONV, black diamonds). Extensor carpi radialis (ECR), extensor digitorum communis (EDC) and extensor carpi ulnaris (ECU) are also shown.

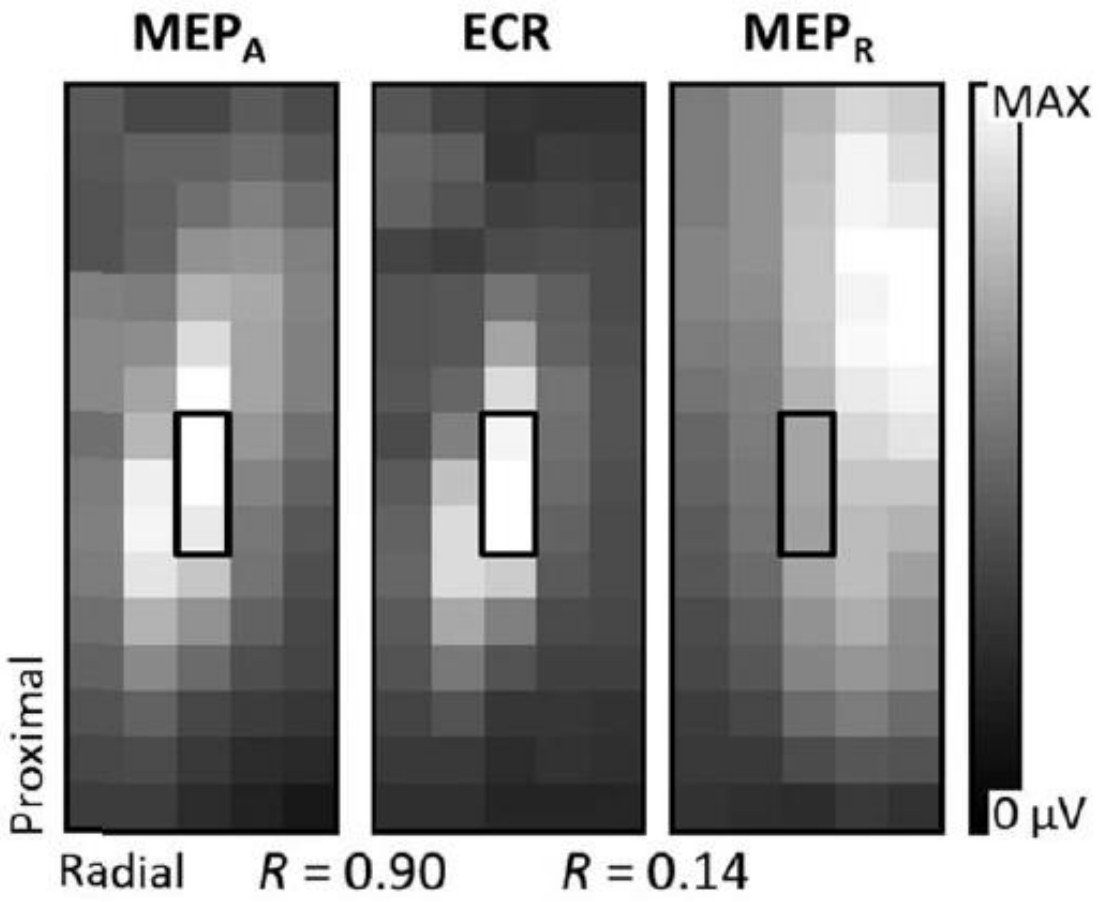


Figure 2: Heatmaps of EMG amplitude distribution during selective activation of the extensor carpi radialis (ECR, middle), and motor evoked potentials while holding a background contraction (MEPsA, left) and at rest (MEPsR, right) of a representative participant. Each map is normalized between 0 and its maximal value. Black boxes indicate the channels with highest amplitude during selective ECR activation. Note how the position of the electrodes that record high EMG amplitude (white/light gray) are similar between ECR and MEPA, but different in MEPR. Correlation values (R) between the selective ECR and the MEPs maps are reported.

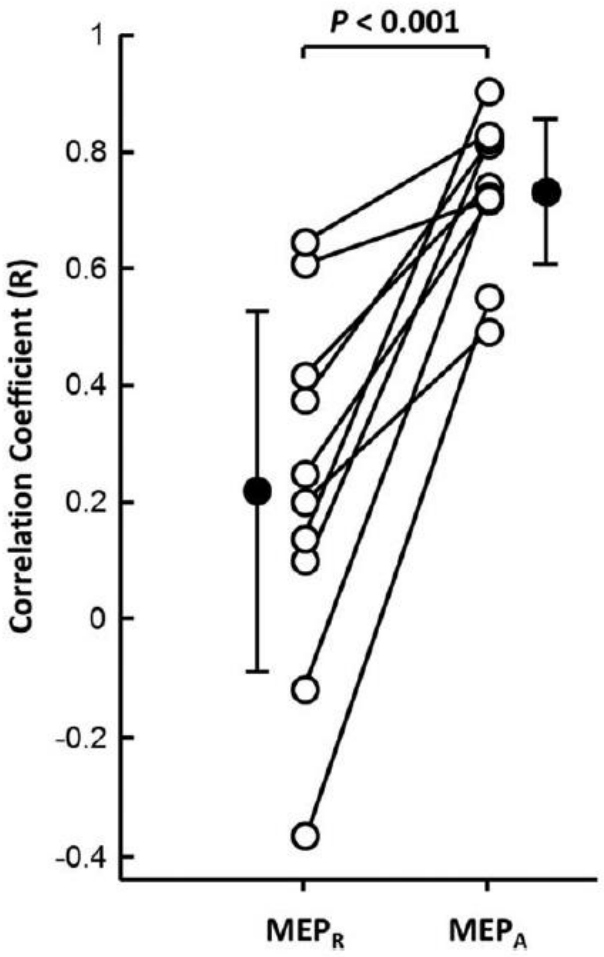


Figure 3: Comparison of motor evoked potential (MEP) selectivity for each participant. High R values describe MEP amplitude distributions similar to those observed during selective extensor carpi radialis activation. The mean R values for the 10 participants are indicated as black dots (bars, standard deviation). MEPs recorded under active conditions (MEPA) were significantly more selective than MEPs recorded at rest (MEPR).

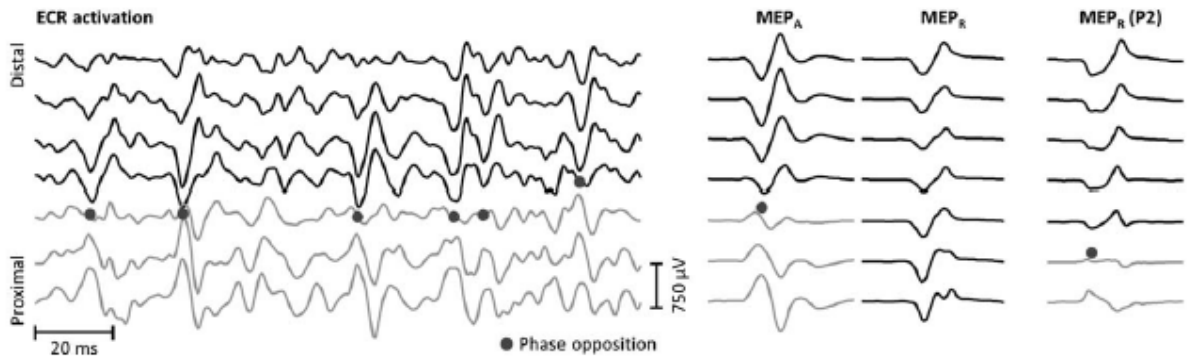


Figure 4: Identification of phase reversal from the virtual bipolar channels calculated from high-density surface electromyography. In the left panel illustrating voluntary extensor carpi radialis (ECR) activation, a channel with low EMG amplitude (dots), and channels with opposite phases proximal and distal to that channel, can be identified in the EMG signals due to the presence of the ECR innervation zone under the electrodes. In the right panel, a similar pattern can be observed in the average MEP under active conditions ($MEP_A$, left column), but not at rest ($MEP_R$, middle column). The potentials in the right column, $MEP_R$ (P2), show the average MEP in the only participant in which an innervation zone could be identified at rest.

**LIST OF ABBREVIATIONS:**

TMS: Transcranial Magnetic Stimulation; EMG: Electromyography; HDsEMG: High-Density Surface Electromyography; ECR: Extensor Carpi Radialis; EDC: Extensor Digitorum Communis; ECU: Extensor Carpi Ulnaris; MEP: Motor Evoked Potential; $MEP_R$: Motor Evoked Potential at Rest; $MEP_A$: Motor Evoked Potential holding a background contraction; M1: Primary Motor Cortex; AMT: Active Motor Threshold; RMT: Resting Motor Threshold; ARV: Average Rectified Value; ICC: Intraclass Correlation Coefficient; SEM: Standard Error of Measurement.