2019

The effect of coil placement and orientation on the assessment of focal excitability in motor mapping with navigated transcranial magnetic stimulation

Reijonen, Jusa

Elsevier BV

Tieteelliset aikakauslehtiartikkelit
© Elsevier B.V.
CC BY-NC-ND https://creativecommons.org/licenses/by-nc-nd/4.0/
http://dx.doi.org/10.1016/j.jneumeth.2019.108521

https://erepo.uef.fi/handle/123456789/7862

Downloaded from University of Eastern Finland's eRepository
The effect of coil placement and orientation on the assessment of focal excitability in motor mapping with navigated transcranial magnetic stimulation

Jusa Reijonen, Laura Säisänen, Mervi Kinnunen, Ali Mohammadi, Petro Julkunen

PII: S0165-0270(19)30378-4
DOI: https://doi.org/10.1016/j.jneumeth.2019.108521
Reference: NSM 108521

To appear in: Journal of Neuroscience Methods

Received Date: 25 June 2019
Revised Date: 26 September 2019
Accepted Date: 12 November 2019


This is a PDF file of an article that has undergone enhancements after acceptance, such as the addition of a cover page and metadata, and formatting for readability, but it is not yet the definitive version of record. This version will undergo additional copyediting, typesetting and review before it is published in its final form, but we are providing this version to give early visibility of the article. Please note that, during the production process, errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

© 2019 Published by Elsevier.
The effect of coil placement and orientation on the assessment of focal excitability in motor mapping with navigated transcranial magnetic stimulation

Jusa Reijonen\textsuperscript{a,b}, Laura Säisänen\textsuperscript{a,b}, Mervi Könönen\textsuperscript{a,c}, Ali Mohammadi\textsuperscript{b}, Petro Julkunen\textsuperscript{a,b}

jusa.reijonen@kuh.fi; laura.saisanen@kuh.fi; mervi.kononen@kuh.fi; ali.mohammadi@uef.fi; petro.julkunen@kuh.fi

\textsuperscript{a}Department of Clinical Neurophysiology, Kuopio University Hospital, P.O. Box 100, FI-70029 KYS, Finland

\textsuperscript{b}Department of Applied Physics, University of Eastern Finland, P.O. Box 1627, FI-70211 Kuopio, Finland

\textsuperscript{c}Department of Clinical Radiology, Kuopio University Hospital, P.O. Box 100, FI-70029 KYS, Finland

Correspondence:
Jusa Reijonen
Department of Clinical Neurophysiology, Kuopio University Hospital, Kuopio, Finland
Address: P.O. Box 100, FI-70029 KYS, Finland
Telephone: +358 40 5052804
E-mail: jusa.reijonen@kuh.fi; jusa.reijonen@gmail.com

Article type: Research Article

Highlights

- Resting motor thresholds increase with increasing distance from the hotspot
- Optimal coil orientation often deviates from the perpendicular-to-sulcus angle
- Motor-evoked potentials may be higher with the coil pointing towards the hotspot
- Electric fields remain high at the hotspot during sulcus-aligned mapping
Abstract

Background: Navigated transcranial magnetic stimulation (nTMS) is used for mapping muscle representations in the primary motor cortex. We used sulcus-aligned mapping and electric field (E-field) modeling to investigate the excitability of the motor hand area for further understanding the methodological limitations of nTMS.

New Method: We studied 10 volunteers to locate the cortical target eliciting the largest responses (the hotspot) in the first dorsal interosseous (FDI) muscle. Six additional targets were placed along the central sulcus at 5-mm distances. Resting motor thresholds (rMTs) and optimal coil orientations were determined at all targets, and a conventional motor mapping was conducted. The cortical E-fields, induced by stimulating the targets with rMT intensities and optimal coil orientations, were modeled in a realistic head geometry to estimate the activated cortical sites.

Results: The rMTs increased with increasing distance from the hotspot ($p<0.001$). The greatest motor-evoked potential (MEP) amplitudes occurred with the coil perpendicular to the sulcus, and with the coil pointing towards the hotspot or the center of gravity of the motor map. The E-field strengths in the hotspot (99±26 V/m) remained above previously estimated thresholds for activation.

Comparison with Existing Methods: Depending on the target location, optimal coil orientations may deviate significantly from the conventional perpendicular-to-sulcus angle. These orientations seem to maintain the E-field stable in the hand knob, regardless of the sulcal shape near the stimulated target.

Conclusions: The coil orientation is crucial for the accuracy of motor mapping, and the apparent motor map may extend due to remote hotspot activation.
**Keywords:** electric field, neuronavigation, neurophysiology, motor cortex, motor-evoked potential, transcranial magnetic stimulation

**Abbreviations:** ADM, abductor digiti minimi; APB, abductor pollicis brevis; BEM, boundary element method; CoG, center of gravity; CSF, cerebrospinal fluid; E-field, electric field; EMG, electromyogram; FDI, first dorsal interosseous; FEM, finite element method; GM, gray matter; MEP, motor-evoked potential; MRI, magnetic resonance image; MSO, maximum stimulator output; nTMS, navigated transcranial magnetic stimulation; rMT, resting motor threshold; TMS, transcranial magnetic stimulation; WM, white matter
1. Introduction

Transcranial magnetic stimulation (TMS) offers a non-invasive method for studying the excitability of the human motor cortex (Barker and Jalinous, 1985). It generates a short current pulse in a coil placed above the scalp, producing a changing magnetic field with lines of flux passing the skull (Hallett, 2000). This induces an electric field (E-field) in the cortex. Depending on the magnitude and direction of the E-field in relation to the cortical structures, corticospinal neurons can be activated, and the resulting response in the contralateral muscles appears as a motor-evoked potential (MEP) in an electromyogram (EMG) (Rossini et al., 2015). This response requires a sufficient stimulation intensity, which is highly individual (Komssi et al., 2004). It is, therefore, often adjusted to the resting motor threshold (rMT), defined as the minimal intensity required to elicit an MEP in a target muscle. In addition to the intensity, the orientation of the coil (which defines the direction of the induced E-field) must be adjusted to optimize stimulation (Säisänen et al., 2008; Schmidt et al., 2015).

The motor hand area is located in a structure of the precentral gyrus, commonly termed the “hand knob” (Yousry et al., 1997). The shapes of the “hand knob” and the adjacent central sulcus exhibit considerable inter-individual variability (White et al., 1997), affecting the identification of individual motor representations. This variability implies that the anatomical accuracy of TMS targeting is crucial (Ahdab et al., 2016). Furthermore, the versatile clinical applications of TMS, such as treatments of chronic pain and depression (Rossini et al., 2015), may also target non-motor areas. This sets additional requirements for the accuracy and repeatability of the targeting methods. Accurate targeting is possible with navigated transcranial magnetic stimulation (nTMS), which utilizes the anatomical information of individual magnetic resonance images (MRIs) (Hannula and Ilmoniemi, 2017). It enables studying the muscle-specific excitability of the motor cortex with higher accuracy (Julkunen et al., 2009) and is suitable for mapping the motor representation areas of
individual hand muscles (Pitkänen et al., 2017). The spatial representation of the mapping results is useful, and quantitative estimation of the areas and centers of gravity (CoGs) can aid in outlining functional motor areas (Borghetti et al., 2008; Julkunen, 2014).

The effects of TMS depend strongly on the orientation of the coil with respect to the underlying anatomy: the induced E-field generates a current in the conductive gray matter, activating optimally oriented corticospinal neurons directly or transsynaptically. This is reflected in varied MEP amplitudes and latencies (Di Lazzaro et al., 2008). By considering the underlying sulcal anatomy, we can adjust the coil orientation during the mapping (“sulcus-aligned mapping”), which improves the capability of revealing within-hand somatotopy (Raffin et al., 2015). However, optimal coil orientation is often assumed to be approximately perpendicular to the central sulcus, which is not always the ideal approach. Balslev et al. (2007) showed that the orientation is within a 45° window around perpendicularity and exhibits broad inter-individual variability. To increase the accuracy of cortical mapping, the optimal orientations should be explored more thoroughly (Bashir et al., 2013). Kallioniemi et al. (2015a; 2015b) elaborated this notion by utilizing nTMS to measure MEP amplitude–coil orientation curves with a wide range of orientations. They observed that a bimodal shape was present in the majority of the curves, indicating that more than one optimal stimulation direction often exists. This may potentially reflect 1) local anatomic arrangement of neural structures or 2) activation of adjacent connected neural structures due to the spread of the TMS-induced E-field. However, the exact reasons for the bimodal shape remain unknown.

To reveal the areas targeted by nTMS, knowledge on the spatial E-field distribution in the cortex is required. The E-field strength in the gray matter is considered as a key factor determining the most likely part of the cortex excited by TMS (Bungert et al., 2017). While the current neuronavigation systems improve the targeting accuracy of TMS and the stability of the induced response (Julkunen et al., 2009), they assume the stimulation site to be located at the cortical...
projection of the coil center or at the local E-field maximum calculated with a simplified head model (Thielscher and Kammer, 2002). More realistic calculation of the TMS-induced E-field distribution is time-consuming and, therefore, currently impractical to implement online during the stimulation. However, applying more accurate models, which consider the individual brain anatomy, improves our ability to characterize the stimulated areas (Thielscher et al., 2011). These models usually rely on structural MRIs to construct a realistic, high-resolution head mesh and use the finite element method (FEM) or boundary element method (BEM) to calculate the E-field distribution in the mesh (Nummenmaa et al., 2013; Windhoff et al., 2013). By combining the realistic E-field distributions with measurements of the MEP, several studies have outlined the likely activation site of the first dorsal interosseous (FDI) muscle in a focal area of the precentral gyrus (Aonuma et al., 2018; Bungert et al., 2017; Laakso et al., 2018; Opitz et al., 2014, 2013).

This study employed nTMS in healthy subjects to investigate the spatial and directional excitability profiles of the FDI muscle by stimulating several targets along the central sulcus. The determination of one-dimensional lateral–medial excitability profiles was combined with a precise estimation of optimal coil orientations and two-dimensional grid-based motor mapping. We aimed at evaluating the importance of nTMS coil placement with respect to the individually varying excitability threshold (rMT) and the effect of anatomical location on the MEP amplitude–coil orientation profiles, thus revealing the optimal coil orientations in relation to individual sulcal anatomy. To locate the likely sites of activation during the sulcus-aligned mapping, we used realistic head models and calculated the E-field distributions induced by stimulating the targets with the rMT intensities and the optimal orientations. These efforts will help us further understand the limitations of nTMS motor mapping and the physical phenomena affecting the accuracy of the resulting maps in evaluating the source of motor activation.
2. Materials and methods

2.1 Subjects and imaging

Ten healthy volunteers (five males, nine right-handed, age: 25–48 years) without contraindications for TMS or MRI participated in the study. The handedness of the participants was based on self-assessment; subject 4 was the only left-handed. All subjects signed a written informed consent, and the local ethics committee approved the study (72/2016). To enable nTMS, structural three-dimensional T1-weighted images were acquired with a 3-T MRI scanner (Philips Achieva 3.0T X, Philips, Eindhoven, The Netherlands) with the following parameters: TR = 8.2 ms, TE = 3.8 ms, flip angle = 8°, voxel size = 1 × 1 × 1 mm. In addition, T2-weighted images were acquired with the following parameters: TR = 2500 ms, TE = 232.7 ms, flip angle = 90°, voxel size = 0.5 × 0.5 × 1 mm.

2.2 Navigated transcranial magnetic stimulation

TMS was performed using an nTMS system (NBS 4.3, Nexstim Plc, Helsinki, Finland) with a figure-of-eight coil, producing single stimulus pulses with a biphasic waveform. By using an integrated EMG device and disposable Ag–AgCl electrodes, we measured EMG with a sampling frequency of 3 kHz from the first dorsal interosseous (FDI), abductor digiti minimi (ADM), and abductor pollicis brevis (APB) muscles of the right hand. The muscle of interest throughout the measurement was the FDI; the signals from the other muscles were monitored to ensure complete relaxation of the hand.

To locate the optimal stimulation target, we first visually determined the approximate location of the hand motor area in the left hemisphere and started stimulating this area. The stimulation intensity was adjusted to produce MEPs with the amplitudes of ~1 mV in the FDI muscle, and the coil was kept tangentially to the scalp. The optimal target, i.e., the hotspot, was determined as the stimulation site repeatedly producing the largest MEPs. To estimate the optimal coil orientation, we
rotated the coil over the hotspot (Julkunen et al., 2009). On average, 59 ± 14 stimuli were applied to locate the hotspot and find the optimal orientation. This coil location–orientation combination was used to determine the rMT of the hotspot by applying a system-integrated threshold hunting paradigm (Awiszus, 2003; Kallioniemi et al., 2017).

We placed six additional stimulation targets along the central sulcus, with three targets on each side (lateral and medial) of the hotspot at approximately 5-mm distances. By considering the individual anatomy, the targets were selected with the aid of visually displayed stimulation grid nodes and numbered in the lateral–medial order from 1 to 6 (Fig. 1A). After the target placement, the coil orientation repeatedly producing the highest MEPs in the FDI muscle was roughly estimated (in steps of ~20°) for each target. This orientation was used in the determination of the target rMT.

To estimate the optimal coil orientations in more detail, we measured the MEP amplitudes as a function of coil orientation at the hotspot and targets 2–5 (Fig. 1B). More precisely, we rotated the coil within ±135° from the optimal orientation with 10 pulses given in each 45° sector (in steps of ~5°), and measured the MEPs at the intensity of 120% of the target rMT (Kallioniemi et al., 2015b). The order of stimulated targets and sectors was randomized. The outermost targets were excluded from these measurements to keep the overall duration within reasonable limits (three to four hours).

Finally, we conducted grid-based motor mapping at the intensity of 105% of the hotspot rMT, starting from the hotspot and extending the area with the aid of a fixed stimulation grid (0.5 cm × 0.5 cm cells) until no responses were obtained (Julkunen, 2014; Pitkänen et al., 2015). Two MEPs were measured at each grid cell with the inter-stimulus interval of 5 s, and the direction of the coil was kept perpendicular to the nearest sulcus during the mapping.
2.3 Motor-evoked potential analysis

The MEPs were automatically detected online with the NBS 4.3 software. Subsequently, the EMG data were visually inspected and analyzed offline, and the responses with peak-to-peak amplitudes of at least 50 μV were accepted as MEPs. To form the MEP amplitude–coil orientation profiles for each target, the MEP amplitudes were smoothed with a 20° moving average window and plotted as a function of coil orientation angle (Fig. 1B). Gaussian functions were fitted to the profiles in the least-squares sense with a custom-made MATLAB script (Kallioniemi et al., 2015b). For the profiles with a bimodal shape, a deconvolution with two Gaussian functions was performed (Kallioniemi et al., 2015a). The targets with two distinct peaks in the bimodal Gaussian fit with normalized amplitudes higher than 0.3 were considered having two separate orientations producing the highest MEPs. This threshold value was considered suitable for clearly distinguishing multiple distinct peaks in the profiles.

The grid-based motor mapping was characterized by calculating its MEP-based CoG:

\[
\begin{align*}
\chi_{\text{CoG}} &= \frac{\sum x_i M_i}{\sum M_i}, \\
y_{\text{CoG}} &= \frac{\sum y_i M_i}{\sum M_i}, \\
z_{\text{CoG}} &= \frac{\sum z_i M_i}{\sum M_i},
\end{align*}
\]

where \(x_i, y_i, \text{ and } z_i\) are the stimulus coordinates in the motor mapping and \(M_i\) is the corresponding MEP amplitude. To outline the mapped motor area, where stimulation produced a motor response, we used the spline interpolation method (Julkunen, 2014); the interpolated coordinates were projected on the gray matter surface for visualization purposes. For visualization of the motor cortical targets and maps, we used MATLAB (version: 2015b, MathWorks Inc., Natick, MA) and GIMP 2.8 (version: 2.8.22, www.gimp.org).
Fig. 1. (color figure, single column) A) A hotspot (HS) and targets 1–6 projected on the cortical surface. B) A normalized and smoothed MEP amplitude–coil orientation curve and a bimodal Gaussian fit (subject 4, target 4). Representative coil targeting plots are visualized for four different coil angles.

2.4 Electric field modeling

To determine the TMS-induced E-fields for the targets in realistic head geometries, we applied the SimNIBS 2.0 software package offline (Thielscher et al., 2015). First, we constructed individual head models from the T1- and T2-weighted MRIs. The models consisted of approximately 500000 nodes and 3500000 tetrahedra, which were classified automatically into five tissue types: white matter (WM), gray matter (GM), cerebrospinal fluid (CSF), skull, and skin. The conductivities of these tissues were chosen to be isotropic and similar to those in previous studies (WM: 0.126, GM: 0.276, CSF: 1.654, skull: 0.010, skin: 0.465 S/m) (Thielscher et al., 2011; Wagner et al., 2004).
The Nexstim figure-of-eight coil was modeled with magnetic dipoles, as described previously by Thielscher and Kammer (2004), to enable fast calculation of its magnetic vector potential in the pipeline. In this method, the figure-of-eight coil is modeled by two circular disks, each disk is divided into rings, and each ring is divided into elements. Then, the dipoles are placed in the center of each element. In this study, the coil parameters, including the number of rings and the inner and outer radii, were estimated from an X-ray image of the coil.

A custom-written MATLAB code was applied to transform the TMS coil locations and orientations from the MRI coordinate system to the SimNIBS coordinate system. The stimulation intensities, measured originally in percentages of maximum stimulator output (%-MSO), were scaled to yield a rate of change of the coil current corresponding to the rMT at each stimulated target. Finally, the FEM was applied to calculate the E-field strength for each element inside the head model. The E-fields were calculated using the coil locations and optimal orientations determined in the nTMS protocol. A custom-written MATLAB script was used to visualize the resulting E-field distribution on the gray matter surface. Specific locations of interest were the cortical projections of the hotspot and targets 1–6, where the E-field strengths were determined. These projections were determined as the closest point on the cortex in the direction from the coil center to the E-field maximum estimated by the neuronavigation system.

2.5 Statistical methods

The effect of stimulation location on the rMT was tested using a repeated measures ANOVA with the target location as an independent variable; the Shapiro–Wilk test was run to test for data normality in the rMT values at each target, and Mauchly’s test was used to test the sphericity assumption. Post-hoc pairwise analyses were conducted, comparing each target with the adjacent targets and the hotspot. The Bonferroni correction was used to correct the p-values for multiple comparisons. Furthermore, the E-field strengths, induced by stimulating the different targets, were
compared at the hotspot and at the targets with the Wilcoxon signed-rank test. The intra- and inter-individual variabilities were compared with Levene's test by using both the rMTs and the E-field strengths; the compared samples were the individual values at the different targets (intra-individual variance) and the values at the hotspot of each individual (inter-individual variance).

To assess the relation between the rMT values and the mapped motor areas, the rMT values were normalized to the rMT value at the hotspot. The targets were classified either inside or outside the motor maps to compare the rMT values between the two classes. The assumptions of normality and homogeneity of variance were tested before comparing the classified rMT values; after testing for normality with the Shapiro–Wilk test, homogeneity of variance was assessed with Levene’s test. Subsequently, the means of the rMT values were compared with an independent samples t-test. The level of significance was $p = 0.05$. MATLAB and SPSS Statistics 23 (IBM Corporation, Somers, NY) were used for statistical analyses.

3. Results

The study protocol was completed successfully in all subjects with the exception of the motor mapping in one subject, which was not completed because of excessive involuntary muscle activity during this phase. The cortical projections of the hotspots and other stimulation targets for individual subjects are presented in Fig. 2.
The cortical locations of the stimulation targets projected on the gray matter surface segmented from the individual MRIs. The red dots indicate the hotspots, and the yellow dots indicate the targets 1–6.
Fig. 3. (color figure, double column) Close-ups for the individual results of the nTMS protocol, showing the stimulated targets with respective rMT values, mapped motor areas, and centers of gravity. The values inside the targets indicate the rMT values (%-MSO) at each target. The optimal coil orientations at the targets are indicated by the arrows, with the length of the shorter arrows indicating the MEP amplitude of the secondary peak in the MEP amplitude–coil orientation profile with respect to the primary peak. The motor mapping of subject 10 was not completed because of excessive muscle artifacts.
3.1 Resting motor thresholds

The summarized individual results of the nTMS protocol are shown in Fig. 3. The rMT values were determined successfully in all subjects and targets; they were normally distributed over individuals in all targets. The assumption of sphericity had been violated ($\chi^2(20) = 36.58, p = 0.021$), and thus a Greenhouse–Geisser correction was used. The repeated measures ANOVA showed a significant effect of target on the rMT value ($F(2.60, 23.38) = 10.69, p < 0.001$). Post-hoc pairwise comparisons revealed that the hotspot had a significantly lower rMT value compared with all targets except target 4, the first medial target from the hotspot (Fig. 4). Target 4 had significantly lower rMT value compared with its adjacent targets 3 and 5. The intra-individual variance in these rMT values ($7 (\%\text{-MSO})^2$) was lower than the inter-individual variance ($45 (\%\text{-MSO})^2$), although the difference was not statistically significant ($p = 0.061$).

![Fig. 4. (single column) Resting motor thresholds. Significant differences with the hotspot are presented with asterisks above the error bars. Significant differences between adjacent targets are presented with asterisks above the horizontal lines. The error bars show the 95% confidence intervals for the group mean values.](image-url)
3.2 Effect of coil orientation

The MEP amplitude–coil orientation profiles showed great intra- and inter-individual variability. The optimal coil orientation was mostly within 45° from the direction perpendicular to the central sulcus (Fig. 3). At many targets, however, the optimal orientation was far from perpendicularity and even aligned with the sulcus. These targets were often the outermost targets (in 5/10 of the subjects), where the optimal orientation was pointing towards the hotspot or CoG of the motor map. Profiles with one peak in the Gaussian fit were the most prominent (38/50 in total), with the widths of the peaks varying considerably between different subjects and targets. Profiles with two clear peaks (12/50 in total) were also observed, especially at the targets closest to the hotspot (Fig. 3). In these profiles, a secondary peak with a lower amplitude was typically detected in addition to the dominant peak.

3.3 Electric fields

The E-fields were the strongest at the gyral crowns. Fig. 5 shows the E-field distributions of subject 4, resulting from the stimulation of the hotspot and targets 1–6. The stimulation of each target, with intensity corresponding to the rMT, yielded high E-field strengths at the projected target location and also at the hotspot (Fig. 6, see Appendix for individual results). These strengths were 106 ± 33 V/m at the targets and 99 ± 26 V/m at the corresponding hotspots. The Wilcoxon signed-rank test showed that the E-field strengths at the hotspot and at the targets differed statistically only when stimulating the outermost targets (Fig. 6). The intra-individual variance in the hotspot E-field values (20 (V/m)²) was lower than the inter-individual variance (711 (V/m)²), although the difference was not statistically significant (p = 0.053).
Fig. 5. (*color figure, 1.5 column*) The E-fields induced on the cortex by stimulating the hotspot (A) and targets 1–6 (B) in one subject (subject 4). The circles (○) show the projected target locations, and the squares (□) show the projected hotspot location. The arrows indicate the optimized coil orientations. The E-fields were calculated with intensities scaled to the resting motor threshold (rMT) at each target.
Fig. 6. (*color figure, single column*) E-field strengths induced at the corresponding targets and at the hotspot when stimulating the different targets. Significant differences between the E-field strength at the hotspot and the E-field strength at the target are presented with asterisks above the error bars. The error bars show the 95% confidence intervals for the group mean values.

3.4 Motor mapping

145 ± 64 stimuli were required to create the motor maps. Individual sizes of the mapped motor areas together with the normalized rMT values and target indices inside the motor maps are shown in Table 1. The normalized rMT values, classified inside or outside the motor maps, were normally distributed in both classification groups. Levene's test indicated that the variances in the groups were significantly different (p < 0.05). Therefore, a t-test for unequal variances was run to compare the means of the groups. The normalized rMT values at the targets outside the motor map (1.16 ± 0.12) were significantly higher than inside the motor map (1.03 ± 0.05) (t(51.97) = 5.78, p < 0.001).
Table 1: Sizes of the mapped motor areas, normalized rMT values, and classification of targets. The bold values indicate the normalized rMT values for the targets inside the motor map.

<table>
<thead>
<tr>
<th>Subject</th>
<th>Mapped motor area (cm²)</th>
<th>Normalized rMT values at targets 1–6</th>
<th>Targets inside the motor map</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>6.90</td>
<td>1.20, 1.13, 1.07, 1.02, 1.13, 1.24</td>
<td>2, 3</td>
</tr>
<tr>
<td>2</td>
<td>4.08</td>
<td>1.27, 1.17, 1.10, 0.97, 0.90, 1.13</td>
<td>3, 4, 5</td>
</tr>
<tr>
<td>3</td>
<td>4.54</td>
<td>1.16, 0.92, 1.00, 1.00, 1.00, 1.16</td>
<td>3, 4, 5</td>
</tr>
<tr>
<td>4</td>
<td>0.52</td>
<td>1.29, 1.35, 1.09, 1.12, 1.32, 1.29</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>7.22</td>
<td>1.07, 1.03, 1.00, 1.00, 1.10, 1.13</td>
<td>1, 2, 3</td>
</tr>
<tr>
<td>6</td>
<td>1.35</td>
<td>1.41, 1.21, 1.05, 1.00, 1.08, 1.05</td>
<td>3, 4</td>
</tr>
<tr>
<td>7</td>
<td>1.62</td>
<td>1.40, 1.28, 1.16, 1.08, 1.16, 1.12</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>1.99</td>
<td>1.00, 1.06, 1.00, 1.00, 1.08, 1.17</td>
<td>4</td>
</tr>
<tr>
<td>9</td>
<td>1.37</td>
<td>1.33, 1.21, 1.08, 1.00, 1.04, 1.08</td>
<td>3, 4, 5</td>
</tr>
</tbody>
</table>

4. Discussion

We investigated the excitability of the primary motor cortex and determined optimal coil orientations by stimulating seven targets along the central sulcus with a wide range of orientations in order to improve our understanding on the limitations for accuracy of the current nTMS motor mapping. As expected, the muscle-specific excitability of the cortex decreased as the distance from the hotspot along the central sulcus increased. The optimal coil orientations were roughly perpendicular to the central sulcus in most targets; however, they also deviated considerably from perpendicularity at many targets, pointing towards the hotspot or CoG of the motor map. Accurate E-field modeling revealed that stimulating the targets with the optimal orientations induced a high, stable E-field at the hotspot, indicating the likely activated area near this location.

The decreasing muscle-specific excitability along the central sulcus (Fig. 4) is in agreement with the measurements conducted by Raffin et al. (2015). Their results show that sulcus-aligned mapping can be applied to obtain medial–lateral excitability profiles by comparing the normalized MEP amplitudes elicited at the intensity of 120% rMT at the different targets. However, intrinsic fluctuations in neural excitability cause large variability in the MEP values (Wassermann, 2002). In addition, the stimulation intensity must remain on the rising phase of the individual input–output
curve to optimally probe the MEP amplitudes (Rossini et al., 2015). Using the rMT values eliminates this dependency on stimulation intensity, providing a more robust measure of excitability. The rMT values still exhibit large inter-individual variability (Fig. 4), which occurs mainly due to differences in corticospinal excitability and coil-to-cortex distance (Danner et al., 2012; Julkunen et al., 2012). However, our results showed that the intra-individual rMT variability, observed in this work due to change of coil position, was lower than the inter-individual variability ($p = 0.061$). Therefore, when tracking changes within individuals, the rMT changes can be much smaller than the inter-individual variability might indicate. The effect of coil-to-cortex distance can be accounted for by modeling the induced cortical E-field, revealing the differences in corticospinal excitability between the subjects.

Despite the large variability in the optimal coil orientations, the optimal orientation was mostly within $45^\circ$ from the posterior–anterior direction perpendicular to the central sulcus (Fig. 3), as suggested by Balslev et al. (2007). The perpendicular orientation was also found optimal in the motor leg area by Richter et al. (2013), who showed that it correlates with the angle of the precentral gyrus. A recent computational study confirmed that this orientation maximizes the E-field strength in the motor cortex (Gomez-Tames et al., 2018). These findings support the use of perpendicular orientation as an approximation if more thorough examination of different orientations is not possible. This orientation maximizes the E-field strength in the nearest gyri (Thielscher et al., 2011), which might indicate activation of the neural structures originating near the targeted cortical site (Fig. 5). Thus, the perpendicular orientation should be applied specifically at the hotspot. At many targets, the optimal coil orientation was not perpendicular to the central sulcus. The optimal orientations seemed rather to point towards the CoG (or the hotspot) of the motor map, regardless of the local shape of the central sulcus (Fig. 3). This increases the E-field strength near the CoG, supporting the existence of a common activation site in a focal area of the “hand knob”. This also emphasizes the importance of considering coil orientation during motor
mapping, as pointing the coil towards the hotspot or the CoG may increase the resulting map size; instead of the targeted cortical site, the hotspot may be activated, and focality of targeting a certain cortical site may suffer.

The MEP amplitude–coil orientation curves often showed a bimodal shape, which was originally observed by Kallioniemi et al. (2015a). They speculated the bimodal shape to result from two separate neuronal populations activated in the different directions. In the current study, the bimodal shape occurred in the targets close the hotspot, and the optimal orientations, obtained from the peaks of the bimodal fit, were close to each other. Thus, it is reasonable to assume that they represent stimulation of optimally oriented subgroups of a spatially compact neuronal population. This could be explained by the direction-sensitive activation of adjacent neurons (e.g., close to the hotspot) due to the spread of the E-field.

The optimal orientations were often close to the lateral–medial or posterior–anterior directions. With monophasic pulses, stimulation in the lateral–medial direction is associated with D-waves, which are produced by direct activation of the proximal parts of the corticospinal neurons in the deeper layers of the motor cortex (Di Lazzaro et al., 2008; Di Lazzaro and Ziemann, 2013). Furthermore, stimulation in the posterior–anterior orientation activates excitatory interneurons in the more superficial layers, resulting in I-waves. However, this association between the coil orientation and the activity of different cortical layers is less consistent when using biphasic stimulation, as in the current study. We chose biphasic stimulation for this study because it is more powerful than monophasic (Kammer et al., 2001) and has been shown to produce MEP amplitude–coil orientation curves with similar shapes as monophasic stimulation (Kallioniemi et al., 2015a). The separation between D-wave and I-wave activities is beyond the scope of this study; however, the relatively strong intensity (120% of rMT) combined with biphasic stimulation increases the occurrence of D-waves together with I-waves (Di Lazzaro et al., 2004). Furthermore, the activation
at different layers of the cortex is dependent on the distribution of the induced E-field with respect to the orientation of the neurons, which should be taken into account when explaining the experimental observations (Laakso et al., 2014; Salvador et al., 2011).

The modeled E-fields were high both at the actual cortical targets and at the hotspot (Fig. 6). The difference in the E-field strengths was statistically significant only when stimulating the outermost targets. The optimal coil orientation was only roughly estimated at these targets, which may cause extra variation in the E-field distribution. However, the E-field strengths at the hotspot were high also when stimulating the outermost targets. Interestingly, the mean E-field strength induced at the hotspot (99 ± 26 V/m) has been shown to be sufficient for motor activation, although the cortical E-field threshold exhibits large inter-individual variability (Julkunen et al., 2012; Mikkonen et al., 2018). This supports the hypothesis, suggested earlier by Thickbroom et al. (1998), that a small region of the cortex is excited even when the coil is moved a substantial distance from the optimal location. The stimulation is less efficient at the further scalp sites; therefore, it requires a higher intensity to include sufficient current density at the site of activation. This phenomenon is nicely demonstrated by comparing Figures 4 and 6: the statistical differences of the motor thresholds are diminished when the E-fields of each target are modeled by using the intensities scaled to the corresponding rMTs, thus revealing the minimum E-fields required for motor activation. These E-fields vary between individuals, reflecting, e.g., the individual differences in corticospinal excitability. The original hypothesis was based on MEP measurements with non-navigated TMS (Thickbroom et al., 1998) and was also suggested by a recent modeling study (Laakso et al., 2018). Our results provide additional evidence by combining the measurements with neuronavigation and E-field modeling. The hypothesis can also be extended to two-dimensional motor mapping: the same focal area is activated when the distance from the map center increases.
if intensity is adjusted to the rMT. Therefore, the motor maps grow larger with increasing intensity (van de Ruit and Grey, 2016).

While the applied nTMS mapping method improves our ability to reveal the somatotopic organization of the motor hand area, the spatial distribution of the induced E-field helps to estimate the excited volume in the cortex. Estimating this volume is still challenging, since it requires setting an excitation threshold for the E-field strength. This threshold also varies spatially depending on the local orientation of the target neurons. By using a global, arbitrary threshold of 150 V/m, Gomez et al. (2013) estimated that a figure-of-eight coil excites volumes of 0.30–0.78% of the head volume, depending on the stimulation depth. In an average head, this would excite volumes in the range of 5–10 cm³. In comparison with TMS, cortical electrical stimulation seems to induce a specific response in a smaller volume of neurons; the excited volume has been estimated to be around 0.01 mm³ in rats (Tsytsarev et al., 2008), and the higher currents applied in the human cortex excite volumes in the range of a few mm³ (Opitz et al., 2014). This difference in the volumes excited by TMS and electrical stimulation can be expected based on the different activation mechanisms: electrical stimulation primarily produces D-waves, while TMS produces I-waves when applied at threshold (Rossini et al., 2015). In TMS, the excited volume likely expands with higher rate when input intensity increases, considering the different approach to inducing the cortical E-field.

The results of the motor mapping (Table 1) showed considerable inter-individual variation in sizes of the mapped motor areas. In addition to the individual variation in the motor representation areas, this variation may increase due to changes in the rMT during the measurement, as the stimulation intensity of the mapping was determined by the initial hotspot rMT measured at the beginning of the nTMS protocol about three hours earlier. The alertness of the subjects may have changed during the measurement, leading to a slightly different rMT at the end of the measurement. This explanation is supported by the normalized rMT values, which show that the
rMT value of the target can predict its location in relation to the motor map, if the normally occurring intra-individual variation in the rMTs is taken into consideration (Koski et al., 2005). In addition, individual cortical morphology affects the extent of E-field spread, which is also reflected in the calculated areas. Due to this E-field spread, the mapped motor areas overestimate the true extent of cortical motor representations. The mapping approach, using a tightly spaced grid (0.5 cm × 0.5 cm cells) and applying two stimuli per cell, was considered suitable for accurately outlining the MEP-eliciting coil locations while keeping the acquisition time reasonable.

The nTMS software uses simplified, spherical models to calculate the induced E-field (Thielscher and Kammer, 2002). These models locate the maximum E-field on the cortex based on a simplified head geometry. This might cause inaccuracies in the locations of the stimulation targets and motor mapping. Currently, realistic head models can only be applied after the measurement to calculate the E-field distribution and locate its maxima. To estimate the activated cortical regions, we analyzed the E-field strengths only near the gyral crowns and not in the sulci, although it is well known that TMS can also stimulate the sulci of the cerebral cortex (Fox et al., 2004). Depending on the stimulation parameters, both of these regions can probably be activated, as suggested by the behavior of D- and I-waves (Di Lazzaro et al., 2008). A recent study by Bungert et al. (2017) suggested that E-field strength is currently the best choice for assessing response-dose dependencies. The best correspondence between the rMT and the E-field strength was observed in regions around the gyral crown, indicating the likely activated region. In addition, the gyral crown is the likely activated region especially when TMS is applied at threshold (Bungert et al., 2017), which was also the circumstance in the E-field modeling of the current study. Since the E-field attenuates strongly when moving deeper in the sulci (Fig. 5), our analysis focused on the regions near the gyral crowns. Increasing the applied stimulation intensity, which was the circumstance in the current study when measuring the MEP amplitude–coil orientation curves, increases the penetration depth of the E-field, thus...
increasing the probability of activation deeper in the sulci. However, also in this case, the E-field strength in the gyri could indicate the approximate location of activation (but not its depth). In future studies, the normal component of the E-field relative to the deeper parts of the sulci should be analyzed in context of suprathreshold stimulation, and E-field modeling could be combined with morphological neuron models to specify the activated neuron populations with higher accuracy (Seo et al., 2016).

5. Conclusions

This study focused on revealing the muscle-specific spatial and directional excitability of the motor hand area by utilizing sulcus-aligned nTMS mapping in order to better understand the limitations associated with nTMS motor mapping. The results demonstrated the importance of using nTMS for optimizing the stimulation parameters, such as the intensity and coil orientation, for more accurate and effective mapping. The modeled E-fields provided supporting evidence for a focal activation site of the FDI muscle in the “hand knob”. They also emphasized critical assessment of the motor map areas, which do not necessarily indicate the activated area on the cortex. However, these maps enable calculating valuable parameters, such as the CoGs, which provide a robust tool for estimating the targeted site and quantifying neuroplastic changes. Visualizing the results together with the anatomical structure is crucial, for example when using the motor mapping in surgical planning.
Acknowledgements

We thank Professor Risto J. Ilmoniemi from Aalto University for comments that greatly improved the manuscript. Jusa Reijonen and Petro Julkunen acknowledge the Research Committee of the Kuopio University Hospital Catchment Area for the State Research Funding (projects 15041763 and 5041771, Kuopio, Finland) and Cancer Society of Finland (Helsinki, Finland). Jusa Reijonen was also funded by the Vilho, Yrjö and Kalle Väisälä Foundation of the Finnish Academy of Science and Letters (Helsinki, Finland).

Competing interests

Petro Julkunen has received travel compensation from Nexstim Plc, manufacturer of navigated TMS systems, and has an unrelated shared patent pending with Nexstim Plc. Laura Säisänen has received unrelated fees for travel expenses from Nexstim Plc. The rest of the authors declare no competing interests.

Appendix

Fig. A.1 shows individual plots of the induced E-fields at the targets and at the hotspot together with the mean curves of Fig. 6.
Fig. A.1. (color figure, 1.5 column) E-field strengths at the corresponding targets (A) and at the hotspot (B) when stimulating the different targets with data for individual subjects.

References


Windhoff, M., Opitz, A., Thielscher, A., 2013. Electric field calculations in brain stimulation based on finite elements: An optimized processing pipeline for the generation and usage of accurate individual head