

1 **Optically pumped magnetometers disclose magnetic field components of the muscular**
2 **action potential**

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21 **Keywords:** peripheral nerve system, muscle action potential, optically pumped magnetometer,
22 finite wire model, magnetic moving dipole model, magnetomyography (MMG),
23 magnetoneurography (MNG)

24 **ABSTRACT**

25 **Aim:** To track the magnetic field generated by the propagating muscle action potential (MAP).

26 **Method:** In this prospective, proof of principle study, the magnetic activity of the intrinsic foot
27 muscle after electric stimulation of the tibial nerve was measured using optically pumped
28 magnetometers (OPMs). A classical biophysical electric dipole model of the propagating MAP
29 was implemented to model the source of the data.

30 **Results:** The signal profile generated by the activity of the intrinsic foot muscles was measured
31 by four OPM devices. Three devices were located above the same muscle to compare the
32 direction and the strength of the magnetic signal while propagating along the muscle.

33 **Interpretation:** OPM devices allow for a new, non-invasive way to study MAP patterns. Since
34 magnetic fields are less altered by the tissue surrounding the dipole source compared to electric
35 activity, a precise analysis of the spatial characteristics and temporal dynamics of the MAP is
36 possible. The classic electric dipole model explains major but not all aspects of the magnetic
37 field. The field has longitudinal components generated by intrinsic structures of the muscle
38 fibre. By understanding these magnetic components, new methods could be developed to
39 analyse the muscular signal transduction pathway in greater detail.

40

41 **What this paper adds:**

- 42 - Technological concepts to record and analyse the small magnetic fields generated by
43 electric muscular activity
- 44 - Model to link the signals measured by the OPM sensors to the underlying physiology
- 45 - Insights into the propagation of muscle action potential and the sequential control of
46 motor activity

47 **Abbreviations:**

- 48 - OPM: optically pumped magnetometer
- 49 - MAP: muscle action potential

50 **1 Introduction**

51 The molecular control of the contractile apparatus of the skeletal muscle is critically
52 dependent on the muscle action potential (MAP) propagating along the muscle fibre after
53 initiation at the neuromuscular endplate (Moritani T, Stegeman D, Merletti R, 2004). After
54 activation of the postsynaptic membrane by acetylcholine (ACh), several different positively
55 charged ions can cross these channels (Martonosi, 2000). The influx of these ions depolarises
56 the membrane of the muscle fibre, the voltage gated sodium channels open and a MAP is
57 generated which propagates along the muscle fibre in both the proximal and distal directions
58 (Farina and Merletti, 2004). In contrast to the neural action potential, the MAP is established
59 by sodium, potassium and calcium ions (Martonosi, 2000). The speed of the MAP is an
60 estimated 4–6 m/s (Farina and Merletti, 2004) slower than the action potentials of motor
61 neurons (around 50 m/s, Ghezzi et al., 1991). Similar to the neuronal action potential (Wiksw
62 et al., 1980), the MAP leads to a specific magnetic field (Zuo et al., 2020), which can be
63 measured by superconducting quantum interference devices (SQUIDs, Reincke, 1993) or
64 optically pumped magnetometers (OPMs) (Broser et al., 2018). However, there are significant
65 anatomical differences between the axon of a neuron and a muscle fibre. First of all, the cross-
66 sectional diameter of a muscle fibre is in the range of 20–100 μm , whereas an axon is typically
67 1 μm in diameter. Further, the membrane of a muscle fibre has a high number of cylindrical
68 infoldings, known as T-tubuli. The MAP is propagated deep in the myocyte along the T-tubuli,
69 in close proximity to the contractile apparatus. The MAP along the T-tubuli generates a radial
70 current that creates an additional magnetic field component in the longitudinal direction of the
71 fibre.

72 The magnetic fields generated by the MAP are in the range of 0.01 to 0.1 nanotesla
73 (nT), and therefore, special devices are required to record these small magnetic fields. OPMs
74 are newly developed sensors (Mhaskar et al., 2012; Colombo et al., 2016; Labyt et al., 2019)

75 that can measure these small magnetic fields. Unlike SQUIDs (Reincke, 1993), OPMs are much
76 smaller and can be placed in close proximity to the muscle.

77 In this study, we developed a measurement setup to record the MAP of the intrinsic foot
78 muscles generated after electric stimulation of the tibial nerve using OPM devices. The
79 magnetic field was measured in two orthogonal directions. The signal-to-noise ratio and
80 reproducibility was tested. Given that magnetic field distributions are geometrically complex,
81 and an intuitive guess of the source of the magnetic field might be misleading, we implemented
82 a classical electric dipole model to explain the sources of the magnetic fields.

83 The model is based on an electric dipole propagating along the muscle fibre. The muscle
84 fibre was approximated as a finite wire, and the MAP was modelled according to the study by
85 Rosenfalck (1969).

86 Given that this model explains some but not all aspects of the magnetic field, a second
87 model based on a moving magnetic dipole was in addition tested. This model is motivated by
88 the estimation of the radial currents inside the muscle as found by Henneberg and Roberge
89 (1997). In comparison to the first model, this model takes into consideration the radial currents
90 resulting from the MAP propagating along the T-tubuli and thus generating a magnetic field
91 component into the longitudinal direction of the muscle fibre.

92 **2 METHOD**

93 *2.1 Recording of the magnetic activity of the MAP*

94 A single-subject, proof of principal study was conducted at the MEG Center of the
95 University of Tübingen in January 2020. The experiment was designed specifically to record
96 the magnetic activity of the intrinsic foot muscles after tibial nerve stimulation.

97 The electric stimulus used to stimulate the nerve interferes with the OPM sensors for
98 about 10 ms. Thus, the stimulation site was chosen such that the time delay between stimulation

99 and muscle activity was greater than 10 ms. Therefore, the tibial nerve was stimulated at the
100 level of the knee.

101 The study aimed to record the activity of the abductor hallucis brevis. This muscle is
102 innervated by the tibial nerve (Figure 1A). The abductor hallucis brevis muscle is located at the
103 medial border of the foot (Figure 1C) and was localised by palpation by an experienced clinical
104 doctor (PB). The muscle inserts on the OS calcaneus. The muscle body is thick and flattens as
105 it stretches forward to the big toe. The muscle ends in a common tendon with the flexor hallucis
106 brevis. This tendon ends on the medial surface of the base of the first proximal phalanx (big
107 toe, hallux). Additional intrinsic foot muscles are located close to the big toe (musculus flexor
108 hallucis and musculus adductor hallucis). These muscles are also innervated by the tibial nerve,
109 and therefore, their MAP could add signal components to the recording (Figure 1C).

110 During the experiment, the tibial nerve was stimulated by two electrodes placed on the
111 skin on the dorsal side of the knee (Figure 1B) at a distance of about 40 cm proximal to the
112 ankle. A series of four OPMs (labelled S, T, U and V) were positioned longitudinal to the medial
113 border of the foot (Figure 1B). The OPM devices were placed such that the z-axis of the sensor
114 pointed towards the plantar side of the foot, and the y axis pointed in the lateral direction (Figure
115 1B). The distance between neighbouring sensors was 3.5 cm. The sensors T, U and V were
116 located above the musculus abductor hallucis and were therefore the main sensors of interest in
117 the study. The participant (a co-author of this paper) gave his informed consent to participate
118 in the study and agreed to the publication of his data. The study was performed in accordance
119 with the Declaration of Helsinki (World Medical Association, 2001).

120 2.2 *Experimental setup*

121 The detailed experimental procedure (Figure 1A,B) began with the subject (a 30-year-
122 old male) lying on a comfortable bed inside of a magnetic shielded room (Ak3b, VAC
123 Vacuumschmelze, Darmstadt, Germany). The subject rested his right leg on a pillow. The right

124 foot was placed in a cardboard box (25×15 cm) filled with rigid foam with a cavity to
125 accommodate the foot. The bottom of the box had four slots to host the OPMs (QZFM-gen-1.5,
126 QuSpin Inc., Louisville, CO, USA). These magnetometers were based on an optically detected
127 zero-field resonance in hot rubidium vapor, which was contained in a vapor cell measuring $3 \times$
128 3×3 mm³. The centre of the cell had a distance of 6.2 mm to the exterior of the housing, which
129 measured $13 \times 19 \times 85$ mm³. This small size allows for easy handling of these sensors and
130 flexibility to adapt the sensors to the specific geometrical situation (Alem et al., 2015; Boto et
131 al., 2017, Osborne et al., 2018). The OPMs are capable of measuring two components of the
132 magnetic field vector: the y and z direction (Figure 1B). They provided a magnetic field
133 sensitivity in the order of 15 fT/ $\sqrt{\text{Hz}}$ in a bandwidth of 3–135 Hz and a dynamic range of a few
134 nT. To adapt to a non-zero magnetic background field, the sensors were equipped with internal
135 compensation coils, which can cancel magnetic background fields with a strength of up to
136 200 nT in the sensing vapor cell.

137 Our OPM measurement system consisted of a total of five OPMs, which simultaneously
138 recorded the magnetic field in two orthogonal directions (y and z). As described above, four
139 sensors were placed inside the slots of the box along a line running across the medial plantar
140 side of the foot (Figure 1C). A fifth sensor was placed just outside the box to measure the
141 magnetic background activity.

142 In order to synchronise the muscular activity with the recording system, the MAP was
143 recorded after electric stimulation of the nerve. To this end, self-adherent electrodes were
144 placed on the skin of the subject in close proximity to the tibial nerve (Figure 1A,B) at the level
145 of the knee. The stimulator (System Plus Fire, Micromed, Venice, Italy) was placed outside the
146 magnetically shielded room, and the two-electrode cables (cathode and anode) were routed
147 through small holes into the magnetic chamber. The tibial nerve was stimulated with a
148 monopolar square wave pulse of 500 μ s duration and an intensity of 35 mA (Broser and
149 Luetschg, 2020).

150 While providing electrical stimuli, a simultaneous trigger signal was sent to the data
151 acquisition system of our MEG system (CTF Omega 275, Coquitlam, BC, Canada). This signal
152 was used to synchronise both the stimulation and the recording. For the recording of the
153 neuromagnetic signal, a sampling rate of 2343.8 samples/s was used. The OPM system
154 delivered two analogue signals for the magnetic field strength in the y and z direction for each
155 sensor. A low-pass filter of 100 Hz was applied to the analogue signal. The OPM system and
156 the analogue digital converter of the recording system both had an intrinsic delay. This delay
157 was measured to be 6.2 ms. The timing of the data was post hoc adjusted so that the stimulus
158 was applied at $t = 0$ s. During the experiment, 10 electric stimuli were applied, and the evoked
159 magnetic muscular activity was recorded. The plots and analyses of the OPM data was
160 performed in R (R Core Team, Vienna, Austria).

161 2.3 Model of the magnetic field of the MAP

162 2.3.1 Geometrical considerations

163 The measurement setup was arranged such that the musculus abductor hallucis was in
164 the xy plane below the OPM sensors T, U and V. Therefore, the position of the MAP on the
165 fibre could be parameterised by:

$$166 \vec{L}(s) = \begin{pmatrix} x_0 + s \cdot \cos(\alpha) \\ y_0 + s \cdot \sin(\alpha) \\ d \end{pmatrix}, \text{ with } \frac{d\vec{L}}{ds} = \begin{pmatrix} \cos(\alpha) \\ \sin(\alpha) \\ 0 \end{pmatrix} \quad (1)$$

167 where $\vec{L}(s)$ is the position pointing to the present position of the MAP; s is the index to
168 parameterise the longitudinal direction of the muscular fibre; x_0 and y_0 are the translation of
169 the fibre in relation to the OPM sensor in the x and y plane, respectively; d is the minimal
170 distance between the OPM sensor and muscle fibre; and α is the angle of the muscle fibre in
171 the xy plane defined by the measurement setup.

172 Without loss of generality the simulation of each sensor was conducted with the
173 assumption that the sensor is located at position (0,0,0). Therefore, for each OPM sensor and

174 each L , the point $\vec{L}(0)$ corresponded to the point of plumb. Figure 2B shows this geometrical
175 arrangement graphically.

176 2.3.2 MAP

177 According to the work by Farina and Merletti (2001), the voltage at the site of the two
178 propagating MAPs can be described by:

$$179 \quad \Delta V_{longitudinal}(s, t) = \psi_1(s - s_{endplate} - v_{AP} * t) + \psi_2(s - s_{endplate} + v_{AP} * t) \quad (2)$$

180 where ψ_1 and ψ_2 being the two MAPs propagating in the proximal and distal directions,
181 respectively. Rosenfalck (1969) described the transmembrane voltage of the MAP using:

$$182 \quad V(s)_{membrane} = \begin{cases} A*s^3 e^{-s} + B; s > 0 \\ 0; s \leq 0 \end{cases} \quad (3)$$

183 where $A = 96 \frac{mV}{mm^3}$ and $B = -90$ mV (Figure 2C). The longitudinal voltage difference can be
184 obtained by differentiation:

$$185 \quad \psi_{1,2}(t) = \left(\frac{dV_{membrane}}{ds} \right) (t). \quad (4)$$

186 The longitudinal voltage gradient $\Delta V_{longitudinal}(s, t)$ was divided by the longitudinal
187 resistance $r_{ix} = 173 \Omega * cm = 17.3 \Omega * mm$ (Henneberg and Roberge, 1997) to obtain an
188 estimate of the longitudinal current. The estimation of the current and current density of the
189 radial current in the T-tubuli was more difficult to obtain. Henneberg and Roberge (1997)
190 created a rigorous mathematical approach and values for discovering the radial current. The
191 sum of the currents in the T-tubuli of a given muscular segment is similar in magnitude to the
192 longitudinal current (Henneberg and Roberge, 1997).

193 2.3.3 Magnetic field of propagating action potentials in general

194 For isolated nerves, the electric field of a propagating action potential generates a
195 magnetic field (Wikswow et al., 1980; Reincke, 1993) with an absolute magnetic field strength
196 of about 70 pT at a distance of about 1 cm from the nerve surface. Wikswow et al. (1980) showed
197 that when the action potential passes the sensor, a characteristic biphasic magnetic signal can
198 be recorded. The turning point of the signal graph correlates with the action potential below the

199 sensor. In a previous study, we recorded the magnetic field of the muscular activity of the
 200 intrinsic hand muscles (Broser et al., 2018). Equation (2) lays the groundwork for the finite
 201 wire model to describe the magnetic field generated by the propagating MAP.

202 2.4 Finite wire model

203 This model assumes that each propagating action potential can be approximated as a
 204 finite wire (Figure 2D-1). Without loss of generality, from here on, only one of the two action
 205 potentials is considered at a time. The magnetic field of this finite wire can be calculated using
 206 the Biot–Savart law (Westgard J., 1997):

$$207 \quad \vec{B}(s) = \frac{\mu_0}{4\pi} \int_s^{s-\Delta s} I(s-t) * \frac{\vec{R}(t) \times \frac{d\vec{L}}{ds}}{|R|^3} dt \quad \text{with}$$

$$208 \quad I(t) = \frac{A_{fibre} *}{r_{ix}} * \left(\frac{dV_{membrane}}{ds} \right) (t), \quad R(s) = \begin{pmatrix} 0 \\ 0 \\ 0 \end{pmatrix}_{\text{PositionOPM}} \quad - \quad L(s) = \begin{pmatrix} -x_0 - s * \cos(\alpha) \\ -y_0 - s * \sin(\alpha) \\ -d \end{pmatrix} \quad (5)$$

209 where s is the index to parameterise the position of the MAP, $\Delta s = 15$ mm and is the longitudinal
 210 extent of the MAP (compare with Equation 3 and Figure 2C), A_{fibre} is the area of electric active
 211 tissue in mm^2 , $r_{ix} = 17.3 \Omega * \text{mm}$ and I is the longitudinal current as calculated from Equation
 212 (3). The sign of I is positive if the movement of positive charges is in the direction of the index
 213 parameter t .

214 Remark: A_{fibre} is dependent on the activated numbers of neuromuscular units and the total
 215 thickness of electric activate muscle tissue.

216 The experimentally determined parameters were A_{fibre} , α , y_0 , x_0 and d . The parameter d
 217 was estimated by muscle ultrasound to be 8 mm. The computations for the finite wire model
 218 were performed using Maple™ (Waterloo, ON, Canada). One simulated example of a resulting
 219 magnetic field generated by an action potential propagated according to Figure 2D-1 is shown
 220 in Figure 2D-2 in the B field in the y direction and Figure 2D-3 in the B field in the z direction.

221 To quantify the residual relative error of the fit, the L^2 norm on the interval -40 to 40 mm off

222 the residual function $\vec{R}(s) := \vec{B}(s) - \overline{OPM}(s)$ was calculated and normalized by the total

223 signal: $\overline{relErr} := \frac{[R]_{L2}}{[OPM]_{L2}}$.

224 2.5 Magnetic dipole model

225 Based on our empiric findings, we developed a second model. This model assumes that
226 the dense T-tubuli network in the muscular fibre generates a net radial current. This current
227 then generates a magnetic dipole. Given the finite expansion of the MAP, we hypothesised that
228 this finite magnetic component can be approximated by a magnetic dipole. The magnetic field
229 generated at the site of the OPM sensor would than calculate to:

$$230 \quad \vec{B}(t) = \frac{\mu_0}{4\pi} \frac{\vec{R}(t) * 3 * (\vec{m} \cdot \vec{R}(t)) - \vec{m} * |\vec{R}(t)|^2}{|\vec{R}(t)|^3} \quad (6)$$

231 with the dipole vector \vec{m} pointing into the direction of the fibre:

$$232 \quad \vec{m} = \begin{pmatrix} \cos(\alpha) * m \\ \sin(\alpha) * m \\ 0 \end{pmatrix} \quad (7)$$

233 where $m[A \cdot m^2]$ is the magnitude of the dipole moment. The experimentally determined
234 parameters were m, α, y_0, x_0 and d . Similar to the finite wire model we simulated the propagation
235 of the muscular action potential along the muscular fibre.

236 3 Results

237 3.1 OPM signal

238 The tibial nerve was stimulated at the level of the knee with 35-mA reliably elicited
239 muscular contractions in the intrinsic foot muscles. As shown in Figure 3, the first signal after
240 stimulation could be detected from the OPM sensors T, U and V about 12 ms after stimulation.
241 In order to estimate the conduction velocity along the tibial nerve, the length of the nerve and
242 the time delay between stimulus onset and the arrival of the signal at the muscle was considered.
243 The lower leg of the subject had a length of 40 cm (knee to medial malleolus), and the distance
244 between the musculus abductor hallucis and the medial malleolus was estimated to be 8 cm.

245 Synaptic transmission typically takes 1 ms. This results in a grossly estimated nerve conduction
246 speed of NCS = $(40 \text{ cm} + 8 \text{ cm}) / (12 \text{ ms} - 1 \text{ ms}) = 0.48 \text{ m} / 11 \text{ ms} = 43 \text{ m/s}$.

247 A total of 10 stimuli were applied to the nerve. The recorded time-dependent magnetic
248 fields for the sensors in the z and y directions are shown in Figure 3. The signal profiles of the
249 individual traces are quite stable and show only moderate variability. The means and standard
250 deviations of the peak-to-peak amplitude of the signals were $0.0094 \pm 0.0021 \text{ nT}$ for sensor S,
251 $0.0648 \pm 0.0145 \text{ nT}$ for sensor T, $0.0759 \pm 0.0051 \text{ nT}$ for sensor U and $0.0496 \pm 0.0029 \text{ nT}$ for
252 sensor V. The background signals were $0.0026 \pm 0.0007 \text{ nT}$ for sensor S, $0.0026 \pm 0.0004 \text{ nT}$
253 for sensor T, $0.0038 \pm 0.0007 \text{ nT}$ for sensor U and $0.0024 \pm 0.0006 \text{ nT}$ for sensor V. The signal-
254 to-noise ratios were therefore 4:1 for sensor S, 24:1 for sensor T, 20:1 for sensor U and 19:1
255 for sensor V. Based on the high signal-to-noise ratio, further analysis of the measured data of a
256 single stimulation was conducted. Wikswa et al. (1980) demonstrated that for the propagating
257 axonal action potential, the magnetic flux shows a change in phase while the action potential
258 passes the sensor. The recorded OPM signals showed a similar bi/triphasic profile (Figure 4)
259 with a zero crossing at 18 ms for sensor S, 20 ms for sensor T, 17 ms for sensor U and 17 ms
260 for sensor V. However, given the larger spatial extent of the MAP, the shape of the magnetic
261 flux signal is more complex.

262 3.2 *Finite wire model*

263 In order to account for the complexity of the MAP signal, a manual parameter search
264 was conducted in order to fit the finite wire model onto the data. Sensors T, U and V were
265 anatomically well-located above the musculus abductor hallucis. The muscles beneath sensor
266 S were less well-defined. Therefore, the analysis focused on the sensors T, U and V. By
267 approximating the velocity of the MAP with $v_{AP} = 5 \frac{\text{m}}{\text{s}}$ (Farina and Merletti, 2004), we could
268 project the time-dependended measured magnetic field strength from the time space into the
269 location space using the formula $z(t) = (t - t_{offset}) \cdot v_{AP}$. The measurement data were

270 transferred into the location space (Figure 4), and a manual parameter search was conducted
271 with the aim to approximate the strength and signal characteristics.

272 For each dataset, the model parameters A_{fibre} , α , y_0 and x_0 were systematically altered,
273 and the signal profiles were visually compared. First the angle α was set to $\alpha = 0^\circ$ or $\alpha = 180^\circ$
274 in order to match the polarity of the first peak in B_y and B_z direction. Second the parameter
275 A_{fibre} was screened to obtain a similar amplitude of the signal in the y direction for all three
276 sensors in consideration (T,U,V). Third the t_{offset} was fine tuned to match the local maxima
277 and minim. Finally, the parameter y_0 was varied to match the magnitude of the B_z component.
278 No further parameters had to be adjusted (see Table 1). In order to quantify the goodness of fit
279 the L2 norm of the residual function normalized by the L2 norm of the signal function was
280 calculated (see Table 2).

281 Interestingly, for sensors T, U and V, only the direction of the propagating action
282 potential (downwards $\alpha = 0^\circ$ or upwards $\alpha = 180^\circ$) and the time offset (t_{offset}) had to be
283 defined. All other parameters were similar for all three sensors. The electric active fibre area
284 A_{fibre} was the same for all three sensors.

285 Based on these parameters, the finite wire model predicted that the MAP propagates beneath
286 sensor T towards the big toe. In addition, it predicted at least one further action potential that
287 propagates in the opposite direction towards the heel and passes sensors U and V. Given that
288 the angles between the two predicted MAPs changed from upwards ($\alpha_T = 180^\circ$) to downwards
289 ($\alpha_{U,V} = 180^\circ$), the MAPs were probably initiated between sensors T and U. This finding
290 directly suggests that the neuromuscular endplate is located between these two sensors.

291 Comparing the signal traces of the recorded (Figure 4, red traces) and the modelled
292 signals (Figure 4, black traces) reveals a relevant difference that cannot be explained by the
293 finite wire model. In the Y direction, the sensors U and V show a triphasic signal trace. The

294 finite wire model only generates a triphasic signal trace if one considers an obscure geometrical
295 arrangement. Triphasic magnetic signal traces are typical for moving magnetic dipoles.

296 We therefore empirically tested if a magnetic dipole model (see Supplementary Figure
297 1) could explain this feature. A parameter search as described above was conducted
298 (Supplementary Table 1) and the signal traces were visualized (see Supplementary Figure 2).
299 As expected, a simulation based on a magnetic dipole returned a triphasic signal trace in the Y
300 direction but a significant angulation of the propagating direction (Supplementary Table 1) was
301 necessary. The residual error for both directions was smaller for the magnetic dipole model
302 when compared to the finite wire model (see Supplementary Table 2).

303 **4 Discussion**

304 In this study, we used OPMs to record the magnetic field generated by the propagating
305 action potential of the intrinsic foot muscles. The MAP was triggered by electric stimulation of
306 the supplying nerve in order to elicit synchronised and timely defined MAP of a high number
307 of simultaneously activated neuromuscular units. The study specifically focused on the
308 musculus abductor hallucis brevis. The three sensors located above this muscle picked up the
309 magnetic component of the MAP with a signal that was greater than 10 times the background
310 activity.

311 As has been previously studied for the nerve AP, we could record a flipping of the B
312 field in the B_y and B_z direction as the MAP passed the sensor. A manual parameter search was
313 conducted to fit a finite wire model to the data. The only one parameter (A_{fiber}) to be fitted to
314 the data was kept the same for all three sensors in consideration. In addition, based on
315 geometrical anatomical considerations the angle of the propagating direction was selected to be
316 either up or downwards. No further adjustments were necessary. The finite wire model could
317 reasonably reproduce the signal traces and the residual error was measure by the L_2 metric. The

318 error was small for the Bz direction but large for the By direction. Based on this model we were
319 able to predict the location of the neuromuscular endplate.

320 Although the model explained major features of the recorded data, the triphasic profile
321 of the measured magnetic fields in the By direction could not be explained by the finite wire
322 model. We therefore empirically tested a magnetic dipole model. This model shows the
323 expected signal profiles and the residual errors are smaller in By and Bz direction, but a
324 significant angulation of the dipole would be required which is not supported by the anatomical
325 situation. Therefore, we hypothesize that a combination of the two approaches would model
326 the magnetic field best. However, to do so the recording of the magnetic field in all three spatial
327 directions is required.

328

329 *4.1 Limitations*

330 Even though the study revealed some very interesting findings, it had some relevant
331 limitation. First the OPM sensor used, only recorded the magnetic field in the Z and Y direction.
332 The field strength in X direction could not be measured and a different model might be obtained
333 when the field in this direction would have been recorded.

334 Second, the study focused on an intrinsic foot muscle. This approach was chosen in
335 order to have a sufficient time delay between electric stimulation and recording. But there are
336 several other muscles in close proximity and so the evaluation of the muscular action potential
337 for the whole extend of the muscle is difficult. In a future study ideally a large muscle with a
338 linear anatomical orientation should be used like the rectus femoris. This muscle would allow
339 to observe the propagation of the muscular action potential over al longer distance. Third, as
340 yet the bandwidth of the OPM sensors is limited and a precise temporal and spatial localization
341 of the signal is restricted. Fourth, the study was based on a stimulated approach. That means
342 the supplying nerve was electrically stimulated in order to record the magnetic field. But for a

343 detailed analysis of the neuro- muscular physiology an approach based on the resting activity,
344 the reflex response or the voluntary activity is necessary. This is especially important to
345 establish this method in clinically medicine.

346 *4.2 Outlook*

347 We are in the process of planning a new set of experiments using the latest generation
348 of OPM devices to record the magnetic field in all three spatial directions. Given the high
349 signal-to-noise ratio shown by the magnetic component of the MAP, we plan to conduct the
350 next series of experiments based on a voluntary muscular activation paradigm or based on the
351 response of the monosynaptic reflex.

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409

410 **Tables**

411 Table 1 – Finite wire model predicted parameters

Sensor	α (°)	y_0 (mm)	A_{fibre} (mm ²)	Offset (ms)
S	77	-4	1	-15.71
T	180	10	2	-17.55
U	0	10	2	-16.30
V	0	10	2	-16.30

412 Note: for all OPM sensors, $x_0 = 0$ and $d = 8$ mm.

413

414 Table 2 – Residual Error - Current Modell

415

Sensor	Y – direction	Z – direction
S	2.558	0.750
T	2.341	0.633
U	0.375	0.287
V	2.112	0.432

416

417

418 **Supplementary Tables**

419 Supplementary Table 1 – Magnetic dipole model predicted parameters

OPM	U	V	T	S	[Unit]
α	120	120	-160	150	[degree]
$l_{polarity}$	1	-1	1	1	
x_0	-0.005	-0.0005	0.001	0.001	[m]
y_0	0	0	0	0	[m]
m_0	$3.5 \cdot 10^{-10}$	$-3.5 \cdot 10^{-10}$	$-3.5 \cdot 10^{-10}$	$-3.5 \cdot 10^{-10}$	$[A \cdot m^2]$
d	0.008	0.011	0.011	0.0145	[m]
v_{Dipole}	5	5	2.5	5	$[\frac{m}{s}]$

420

421 Supplementary Table 2 – Residual Error - Magnetic Dipole Modell

422

Sensor	Y – direction	Z – direction
S	0.618	1.44
T	0.428	0.464
U	0.417	0.336
V	1.397	0.304

423

424 **Figure legends**

425 **Figure 1:** Principal setup of the experiment. (A) Schematic anatomical drawing of the lower
426 leg. The black circle at the level of the knee highlights the area on the skin where the tibial
427 nerve was stimulated. (B) Schematic drawing of the lower leg. Lateral view onto the medial
428 aspect of the lower leg together with the relevant structures and technical equipment. (B1)
429 Location of the self-adherent stimulation electrodes (black = cathode, red = anode). (B2) Tibial
430 nerve. (B3) Intrinsic foot muscles, specifically the musculus abductor hallucis. (B4) Four OPM
431 recording devices. Coordinate system in the upper right. The y axis points out of the drawing
432 area. (C) Position of the four OPM devices (S, T, U and V) in relation to the anatomical
433 structure. (C1) Musculus abductor hallucis. (C2) Musculus flexor hallucis brevis. (C3)
434 Musculus adductor hallucis. The x axis points from the big toe to the heel. The y axis points
435 from digit V to digit I.

436
437 **Figure 2:** Illustration of the methodological considerations. (A) Schematic drawing of a muscle
438 (dark red) and axon of a motor neuron (green) with the motor endplate. Two (v), (-v) action
439 potentials are shown propagating from the endplate to the distal ends of the muscle fibres. The
440 action potential first generates a current with a technical current direction (movement direction
441 of the positive charges) in the direction of the moving action potential. Then, during the
442 repolarisation phase, a current with a technical current in the opposite direction of the
443 propagating action potential is generated. Both currents generate a circular magnetic field in
444 the direction as defined by the right hand rule. (B) Geometrical situation of OPM device (grey),
445 muscle fibre (dark red) and propagating MAP (grey circle with the currents I and movement
446 velocity V). In the figure, d is the shortest distance between the sensor and muscle, l is the
447 longitudinal position of the action potential in relation to the *Lotfußpunkt* (point of shortest
448 distance d to the OPM sensor) and R is the vector connecting the OPM sensor and position of

449 action potential with the components. The origin of the coordinate system is defined by the
450 position of the OPM sensor.

451
452 **Figure 3:** Recorded time-dependent magnetic field strength for each OPM sensor (S, T, U and
453 V). (A) Anatomical location of the four OPM sensors (same as Figure 1). (B) Plots of the
454 recorded time-dependent magnetic field strength in the y (left) and z (right) directions for each
455 sensor S, T, U and V (top to bottom). A total of 10 measurements were performed. Each
456 recorded trace is shown as a fine grey line. The first trace – which was used for further analysis
457 – is shown in black. The small horizontal grey lines shown on the plots for the z direction in
458 the time window 5–10 ms correspond to the minimum/maximum of the signal during this
459 period. These maxima were used to approximate the background signal. The red/blue open
460 circles mark the maximum/minimum of the biphasic peaks. The green line on each graph of the
461 y direction marks the second local maximum/minimum. The green line on the graphs of the z
462 direction marks the turning point of the graph. This point corresponds to the time when the
463 action potential passes the *Lotfußpunkt* (point of shortest distance d to the OPM sensor).

464
465 **Figure 4:** Comparison of experimentally recorded and modelled magnetic field. For each OPM
466 sensor (S, T, U and V), the recorded magnetic field strength in the y and z direction (red trace)
467 is shown. Based on the finite wire model, the hypothetical sources were modelled, and the
468 corresponding magnetic field strengths were plotted (black traces). The plots on the far right
469 show the presumed offset in the y direction, the longitudinal direction in the xy plane of the
470 muscular fibre (black line) and the direction of the MAP. Note the change in the direction of
471 the MAP between sensors T and U. The MAP likely originated between these two sensors.

472
473
474

475 **Supplementary Figure 1:**

476 Similar Figure as Figure 2 but with the illustration of the magnetic dipole model. Inset in Panel

477 A illustrates how the radial current along the T – tumuli could generate a magnetic dipole.

478 Graphs in Panel C show the signal profile of a simulated moving magnetic dipole.

479

480 **Supplementary Figure 2:**

481 The plot on the left and in the middle show the measured signals for the sensors S, T, U and V

482 top to bottom (red lines). The black line is the result of the simulation based on the magnetic

483 dipole model and model parameters described in the Supplementary Table 1.

484 The axis of abscissae states the distance of the dipole from the “Lotfußpunkt” (point of shortest

485 distance d to the OPM sensor) in cm. The axis of ordinate shows the magnetic field in nano

486 Tesla.

487 The plots on the far right show a two dimensional coordinate system of the xy plane with the z

488 direction point out of the plane and the OPM sensor at the position $(x=0, y=0, z=d)$.

489 The redline on these plots show the linear path of the moving dipole. The blue arrow on the

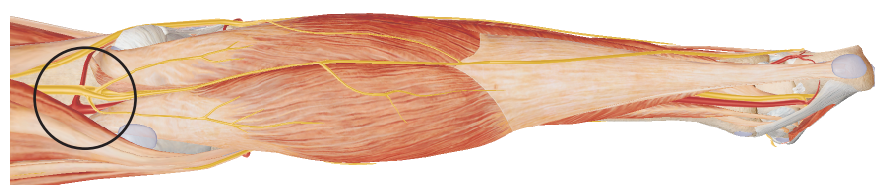
490 redline points into the direction of movement in relation to time. The red arrow at the centre of

491 origin $(x=0, y=0)$ shows the direction of the magnetic dipole.

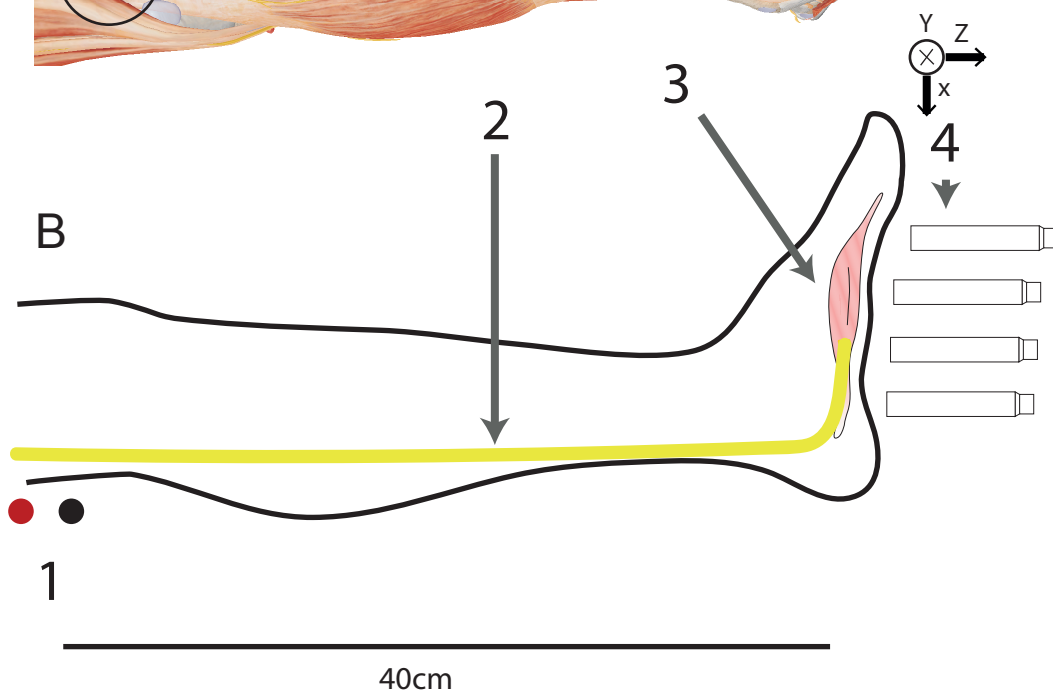
492

Figure 1

A



B



C

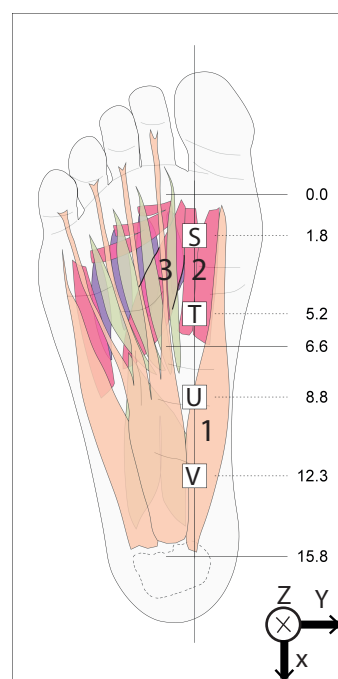


Figure 2

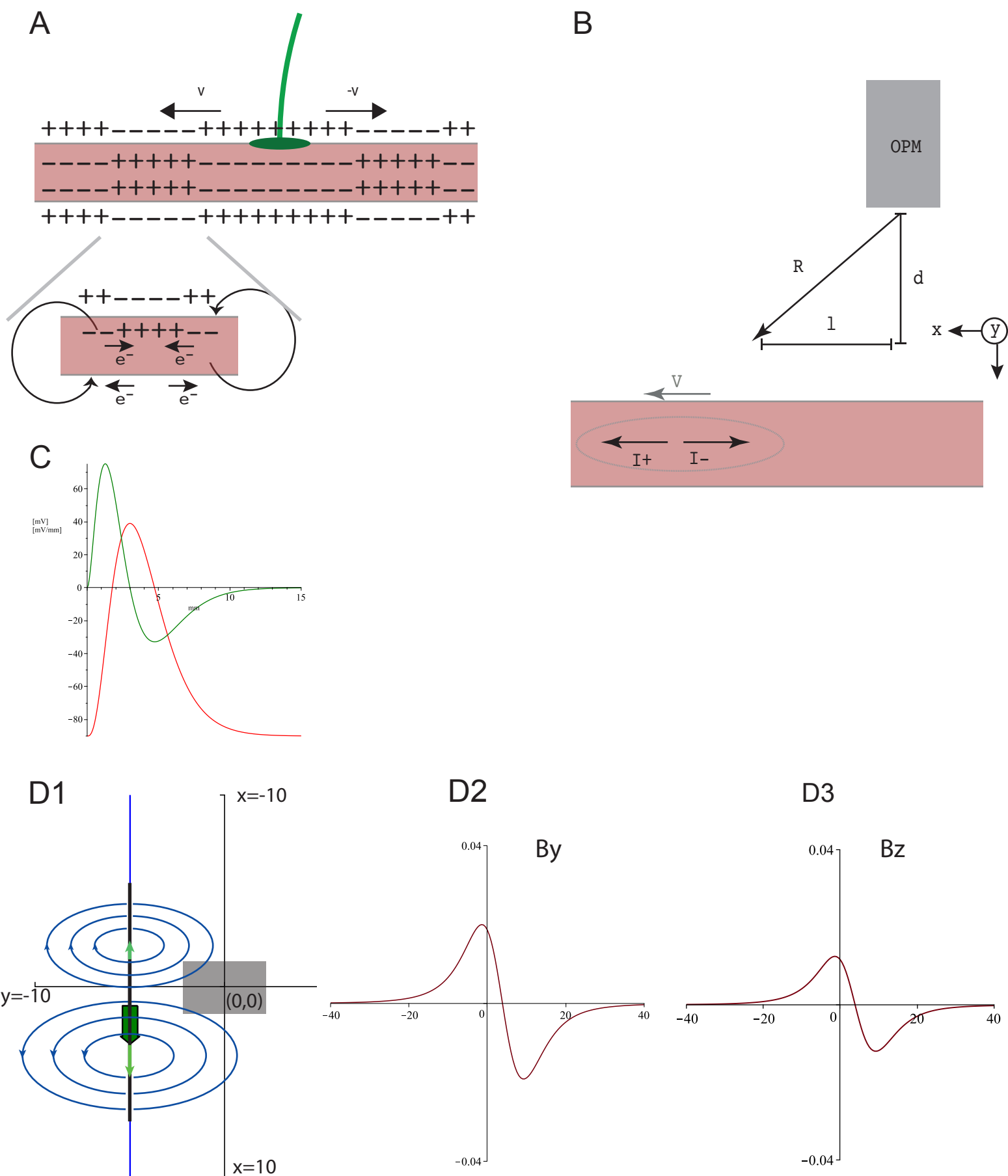


Abbildung 3

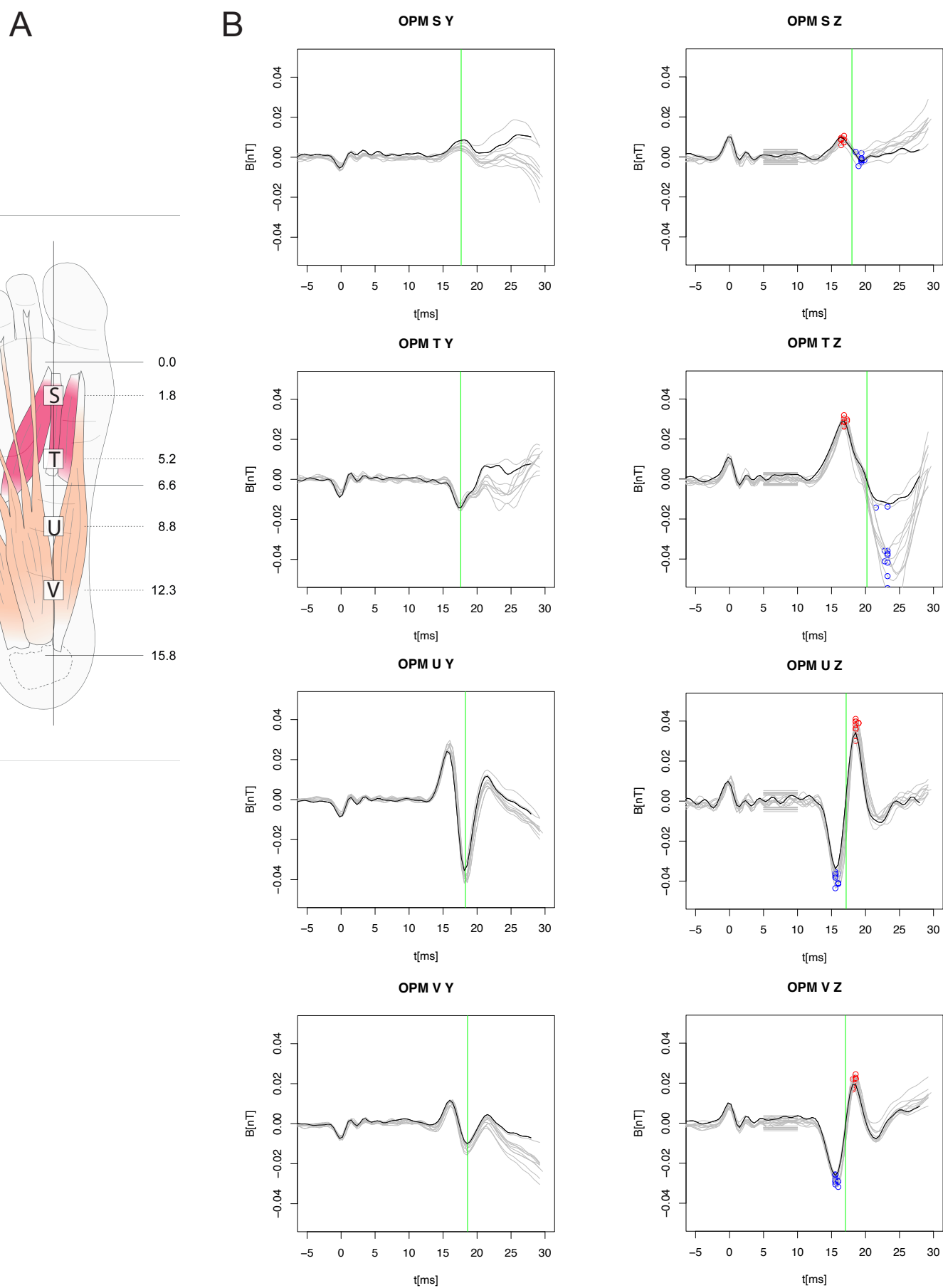
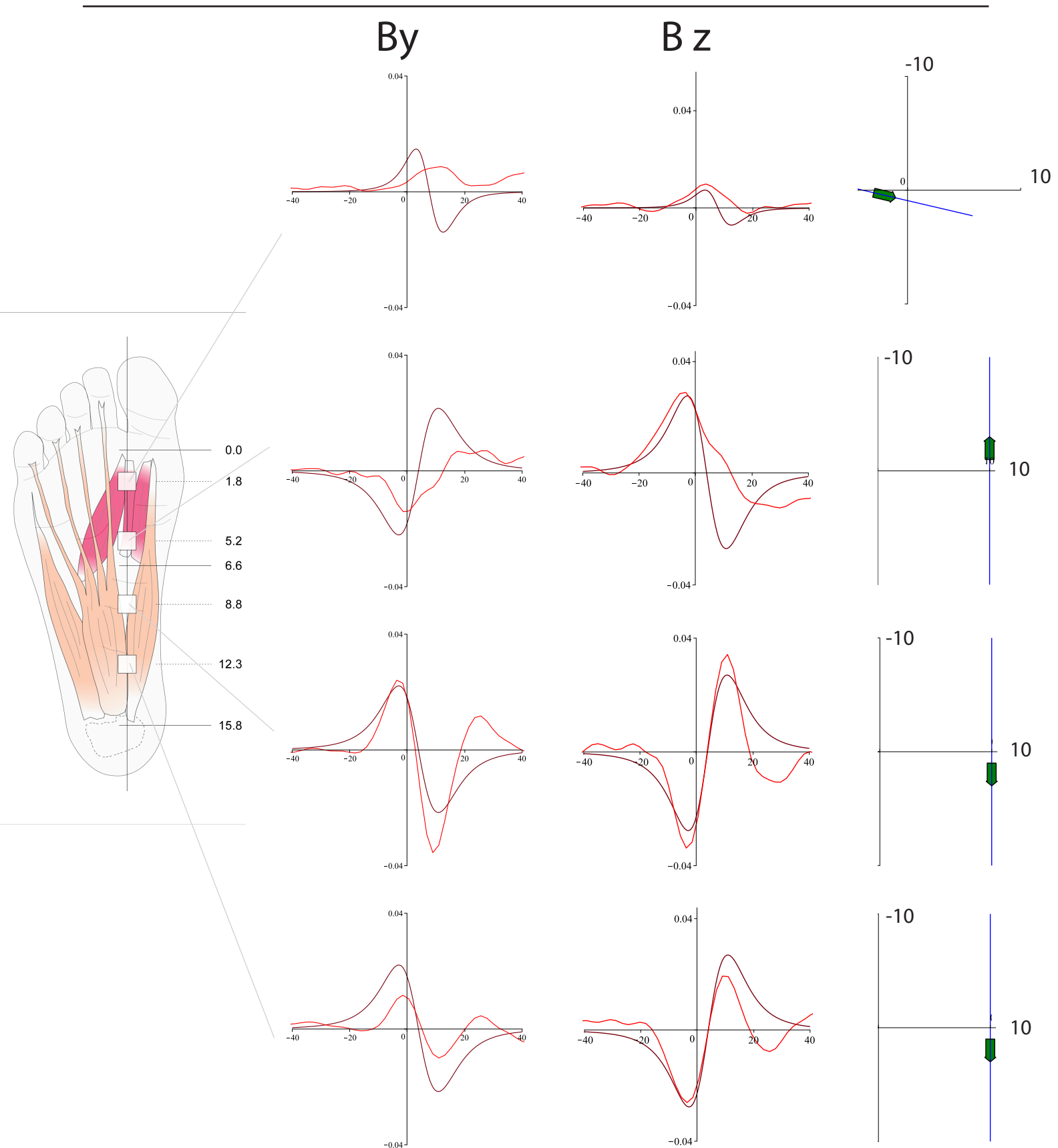
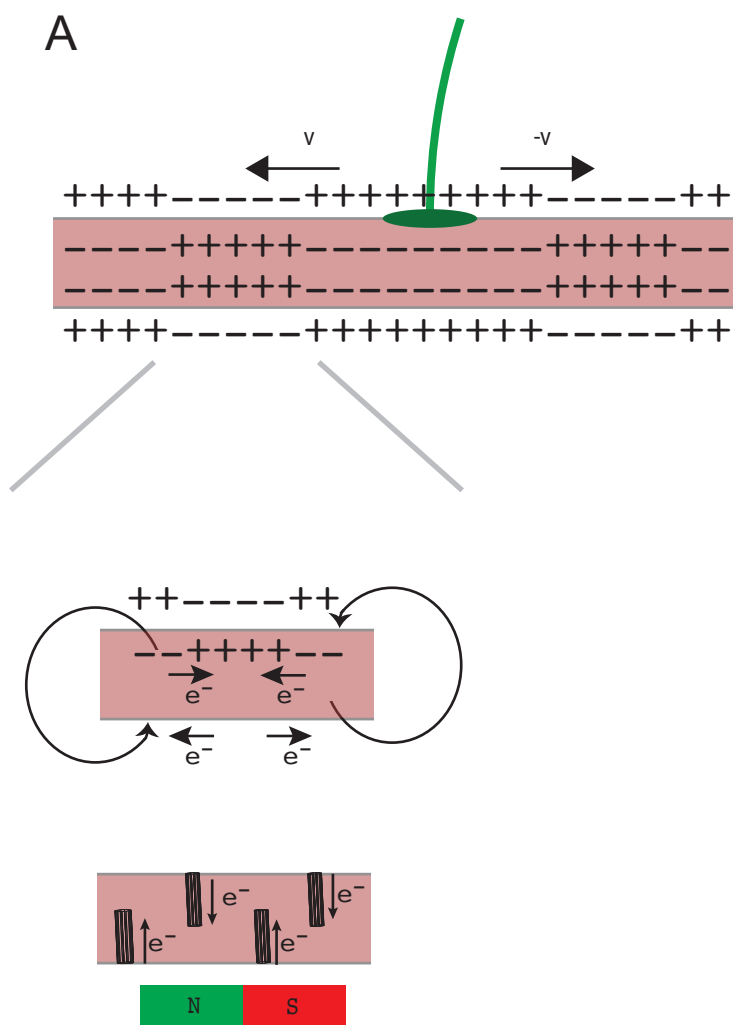


Figure 4

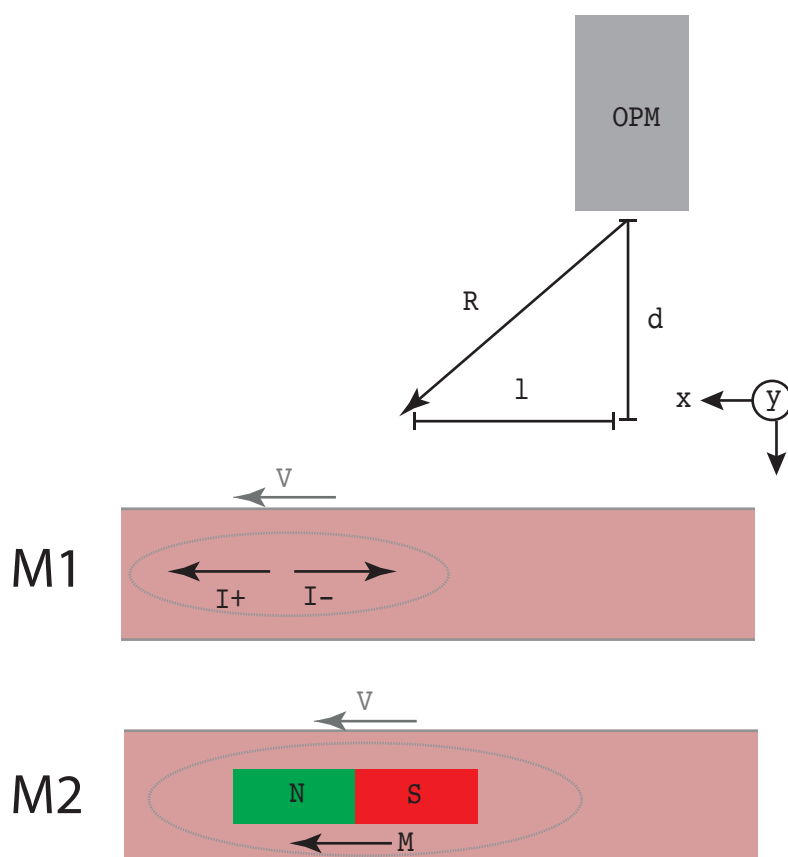


Supplementary Fig. 1

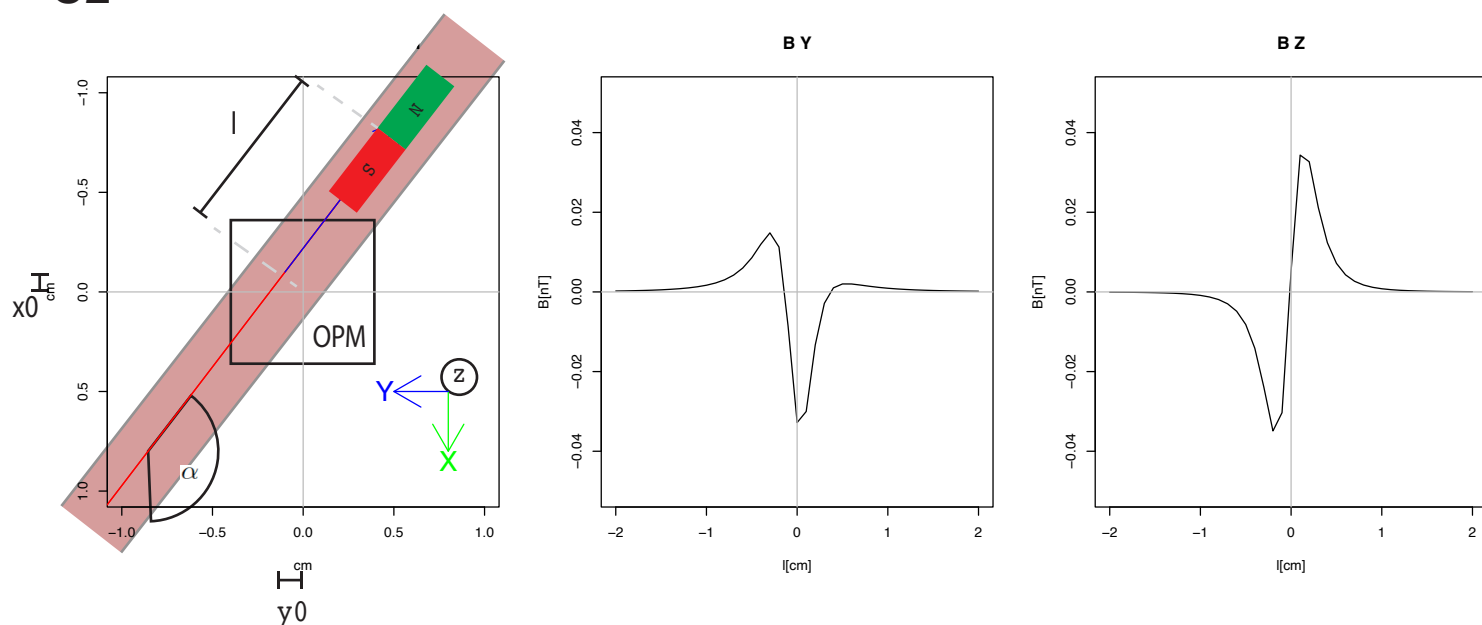
A



B



C2



Supplementary Fig. 2

