Supplementary material

Supplementary table 1. Summary of MRI acquisition parameters.

Sequence	FOV [mm]	Voxel size [mm]	Rec voxel size [mm]	TE [ms]	TR [ms]	TI [s]	FA [°]	MoCO	SENSE	Scan time [min]
3D[a] MPRAGE	228 x 228 x 166	0.65 iso	0.65 iso	3.2	7.5	NA	8	Yes	AP: 1.6, RL: 1	7:13
3D MP2RAGE[c]	205 x 205 x 192	0.85 x 0.85 x 0.8	0.8 iso	3.5	8.0	404/3004	6/5	Yes	AP: 1.8, RL: 1.8	11:39
3D FLAIR	230 x 230 x 168	0.7 iso	0.69 x 0.69 x 0.7	391	5000	1.83	55	Yes	AP: 2, RL: 1.5	11:10
3D T2w-TSE[d]	256 x 256 x 190	0.8 iso	0.4 iso	319	3719	NA	100	Yes	AP: 2, RL: 2	11:54
3D T1w	246 x 246 x 174	0.99 x 1 x 1	0.85 x 0.85 x 1	2.2	5.0	NA	7	No	AP: 2, RL: 2.5	01:55
MS DTI-EPI[b,e]	200 x 200 x 121	1.79 x 1.85 x 1.8	1.79 x 1.79 x 1.8	54	13935	NA	90	No	AP:4	8:08

[a]All 3D data were collected with sagittal read-out,
[b]Multi-slice DTI data were acquired axially.
[c]The overall MP2RAGE TR was 5.5s, and data were acquired with 0.775 halfscan along y, TFE factor 128.
[d]The T2w-TSE factor was 200.
[e]The DTI acquisition used a single b-value of 1000 over 32 gradient diffusion encoding directions.
Abbreviations: FOV = field-of-view, rec = reconstructed, TE = echo time, TR = repetition time, TI = inversion time, FA = flip angle, MoCo = prospective motion correction, SENSE = sensitivity encoding (parallel acceleration factors), AP = anterior-posterior, RL = right-left, iso = isotropic, MS = multi-slice, EPI = echo planar imaging,

Supplementary table 2. Demographics and neurophysiological results

	Dominant				Non-dominant			
	HC (N=20)	CL- (N=18)	CL+ (N=19)	All patients (N=37)	HC (N=20)	CL- (N=16)	CL+ (N=21)	All patients (N=37)
Demographics and clinical information[a]								
Age years mean (SD)	43.2 (13.0)	43.9 (10.9)	49.2 (12.1)	46.6 (11.7)	43.2 (13.0)	41.7 (10.4)	50.4 (11.4)	46.6 (11.7)
Sex n (% female)	14 (70.0%)	14 (77.8%)	11 (57.9%)	25 (67.6%)	14 (70.0%)	14 (87.5%)	11 (52.4%)	25 (67.6%)
Phenotype n (% RRMS)	-	16 (88.9%)	10 (52.6%)	26 (70.3%)	-	15 (93.8%)	11 (52.4%)	26 (70.3%)
EDSS median [range]	-	3.25 [0, 6.5]	3.5 [1.5, 6.5]	3.5 [0, 6.5]	-	3.0 [0, 6.5]	4.0 [1.5, 6.5]	3.5 [0, 6.5]
FS pyramidal median [range]	-	1.0 [0, 3]	3.0 [0, 4]	2.0 [0, 4]	-	1.0 [0, 3]	3.0 [1, 4]	2.0 [0, 4]
Missing (*n*)	-	0	1 (5.3%)	1 (2.7%)	-	0	1 (4.8%)	1 (2.7%)
FS sensory median [range]	-	1.0 [0, 3]	2.0 [0, 3]	2.0 [0, 3]	-	1.50 [0, 3]	2.0 [0, 3]	2.0 [0, 3]
Missing (*n*)	-	0	1 (5.3%)	1 /2.7%)	-	0	1 (4.8%)	1 (2.7%)
Disease Duration years median [range]	-	8.5 [1, 25]	16 [3, 45]	10.0 [1, 45]	-	8.5 [1, 26]	14.0 [1, 45]	10.0 [1, 45]
Motor Fatigue median [range]	-	20.5 [0, 31]	21.0 [6, 38]	21.0 [0, 38]	-	20.5 [0, 38]	21.0 [6, 38]	21.0 [0, 38]
Neurophysiological measurements[a]								
N20 latency ms mean (SD)	22.6 (1.92)	24.1 (4.49)	27.1 (8.68)	25.6 (6.97)	22.7 (1.89)	22.0 (1.23)	29.5 (9.78)	26.2 (8.17)
Missing (*n*)	0	0	1 (5.3%)	1 (2.7%)	0	1 (6.2%)	2 (9.5%)	3 (8.1%)
RMT % mean (SD)	33.8 (5.97)	33.3 (8.44)	38.3 (10.2)	35.9 (9.60)	34.9 (6.41)	32.1 (7.63)	40.9 (8.48)	37.1 (9.15)
MEP_{max} % mean (SD)	54.8 (13.6)	46.7 (17.3)	36.1 (17.6)	41.3 (18.0)	57.3 (12.4)	55.1 (15.0)	35.3 (22.2)	43.9 (21.6)
CMCT ms mean (SD)	5.36 (0.921)	7.01 (3.27)	10.1 (8.50)	8.58 (6.60)	5.15 (0.828)	6.19 (2.73)	10.7 (9.40)	8.76 (7.57)

[a]Data is from the cohort that participated in the neurophysiological examination (N=57).
WM = white matter, SD = standard deviation, RMT = resting motor threshold, MEP = motor evoked potential, CMCT = corticomotor conduction time, HC. = healthy control.

Supplementary table 3. Correlations with SM1-HAND total and cortical lesion subtype number and volume

	Composite sensory score	Composite motor score	CMCT	MEP_{max}
Count[a]				
Total cortical lesions	-0.01 (p=1)	-0.18 (p=1)	-0.19 (p=1)	-0.04 (p=1)
Type I	-0.11 (p=1)	0.00 (p=1)	-0.15 (p=1)	0.26 (p=0.78)
Type II	0.13 (p=1)	-0.00 (p=1)	-0.13 (p=1)	-0.18 (p=1)
Type III/IV	0.00 (p=1)	-0.04 (p=1)	0.05 (p=1)	-0.12 (p=1)
Volume[a]				
Total cortical lesions	0.20 (p=1)	-0.03 (p=1)	0.06 (p=1)	-0.16 (p=1)
Type I	-0.06 (p=1)	0.04 (p=1)	-0.07 (p=1)	0.18 (p=1)
Type II	0.18 (p=1)	0.04 (p=1)	-0.17 (p=1)	-0.12 (p=1)
Type III/IV	-0.00 (p=1)	-0.05 (p=1)	0.08 (p=1)	-0.11 (p=1)

[a]Data is from CL+ patients only.
CMCT = corticomotor conduction time, MEP = motor evoked potential

Supplementary table 4. Mixed linear models using the paracentral control region of interest

Main effects[a]		**Post hoc comparisons**[b]				
Composite motor score						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$22.11_{(2)}$	CL- – HC	1.72	0.35	4.92	**<0.001**
P-value	**<0.001**	CL+ – HC	1.61	0.43	3.741	**<0.001**
		CL+ – CL-	-0.11	0.37	-0.3	0.77
Composite sensory score						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$25.14_{(2)}$	CL- – HC	1.09	0.21	5.13	**<0.001**
P-value	**<0.001**	CL+ – HC	1.17	0.27	4.43	**<0.001**
		CL+ – CL-	0.08	0.23	0.34	0.73
CMCT						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$9.12_{(2)}$	CL- – HC	0.28	0.12	2.38	**0.03**
P-value	**0.01**	CL+ – HC	0.40	0.13	3.09	**0.006**
		CL+ – CL-	0.12	0.09	1.37	0.17
MEP_{max}						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$11.49_{(2)}$	CL- – HC	-9.89	4.54	-2.18	0.06
P-value	**0.003**	CL+ – HC	-17.89	5.15	-3.47	**0.002**
		CL+ – CL-	-8.01	3.94	-2.03	0.06
RMT						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$2.21_{(2)}$	CL- – HC	-	-	-	-
P-value	0.33	CL+ – HC	-	-	-	-
		CL+ – CL-	-	-	-	-
N20 latency						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$6.44_{(2)}$	CL- – HC	$1.31e^{-04}$	$5.27e^{-05}$	2.48	**0.04**
P-value	**0.04**	CL+ – HC	$5.51e^{-05}$	$6.07e^{-05}$	0.91	0.36
		CL+ – CL-	$-7.56e^{-05}$	$5.06e^{-05}$	-1.49	0.27

Significant tests are highlighted in bold.
[a]Likelihood-ratio tests of a main effect of group. P-values are uncorrected.
[b]Tukey's post hoc tests. P-values are corrected using the Holm-method within each model.
HC = healthy control.

Supplementary table 5. Mixed linear models using the caudal middle frontal control region of interest

Main effects[a]		**Post hoc comparisons**[b]				
Composite motor score						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$23.18_{(2)}$	CL- – HC	1.56	0.36	4.37	**<0.001**
P-value	**<0.001**	CL+ – HC	1.89	0.38	4.95	**<0.001**
		CL+ – CL-	0.33	0.31	1.08	0.28
Composite sensory score						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$25.19_{(2)}$	CL- – HC	1.08	0.22	4.98	**<0.001**
P-value	**<0.001**	CL+ – HC	1.16	0.24	4.92	**<0.001**
		CL+ – CL-	0.08	0.19	0.41	0.68
CMCT						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$7.83_{(2)}$	CL- – HC	0.30	0.12	2.52	**0.02**
P-value	**0.02**	CL+ – HC	0.34	0.12	2.88	**0.01**
		CL+ – CL-	0.05	0.07	0.69	0.49
MEP_{max}						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$8.74_{(2)}$	CL- – HC	-10.72	4.56	-2.35	**0.04**
P-value	**0.013**	CL+ – HC	-14.31	4.65	-3.08	**0.006**
		CL+ – CL-	-3.59	3.26	-1.10	0.27
RMT						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$1.57_{(2)}$	CL- – HC	-	-	-	-
P-value	0.46	CL+ – HC	-	-	-	-
		CL+ – CL-	-	-	-	-
N20 latency						
			Estimate	Std. error	z-value	P-value
$\chi^2_{(df)}$	$4.31_{(2)}$	CL- – HC	-	-	-	-
P-value	0.12	CL+ – HC	-	-	-	-
		CL+ – CL-	-	-	-	-

Significant tests are highlighted in bold.
[a]Likelihood-ratio tests of a main effect of group. P-values are uncorrected
[b]Tukey's post hoc tests. P-values are corrected using the Holm-method within each model.
HC = healthy control

Supplementary methods

Neurophysiological recordings

Somatosensory evoked potentials

SEPs were recorded over the somatosensory cortex of each hemisphere individually. Electrical stimulation was applied to the index finger of the right and left hand, over the proximal interphalangeal joint using a bipolar electrode montage. We applied 500 square pulses of 200 μs duration at a frequency of 2 Hz, at 300% of the perceptual threshold determined to the nearest 0.5 mA, using a constant current generator (Digitimer, Cambridge Electronic Design, Cambridge, UK). A recording EEG electrode was placed on CP3 and CP4 and a reference electrode over Fz. A ground electrode was positioned on the left earlobe. Responses were filtered (bandpass, 5-2000 Hz) and amplified (factor 100.000, Digitimer, Cambridge Electronic Design, Cambridge, UK), digitalized (1201 micro Mk-II, Cambridge Electronic Design, Cambridge, UK, sampling frequency 5000 Hz), recorded using Signal (Cambridge Electronic Design, Cambridge, UK) and stored for later off-line analysis.

Surface EMG

Electrical muscle activity was recorded from the left and right first dorsal interosseous (FDI) muscles using surface EMG electrodes (Ambu, Neuroline 700, Ballerup, Denmark) placed in a bipolar belly-tendon montage. Using the same setup as for EEG, the analog EMG signal was bandpass filtered, amplified (factor 200-1000), digitalized, recorded using Signal and stored for later off-line analysis. During recordings, background EMG was carefully monitored by the experimenter, and participants were instructed to relax whenever background EMG exceeded noise levels.

TMS procedures

Neuro-navigated Transcranial magnetic stimulation (TMS)

TMS was applied over the hand-knob area of the left and right hemisphere. Stimulations were delivered using a MagPro x100 Option stimulator (Magventure, Skovlunde, Denmark) connected to a MC-B70 figure-of-eight coil. The coil was placed tangentially over the scalp,

with the handle pointing in approximately 45° angle to the mid-sagittal plane. Biphasic pulses with an AP-PA current induction in the brain were used to elicit motor evoked potentials (MEPs). The motor hotspot for the right and left FDI was determined through a mini-mapping procedure using supra-threshold stimulations and defined as the site eliciting the largest and most consistent MEPs. Stereotactic MRI-based neuro-navigation (Localite, Sankt Augustin, Germany), was used to mark the hotspot and monitor coil position throughout the experiment. Individual 7T T1-weigthed images were used for automated brain surface reconstruction and participants were registered into the system using three focal points (nasion, left and right ear) along with a minimum of 150 surface registration points. Resting motor thresholds (RMT) were determined to the nearest 1% of maximum stimulator output and defined as the minimal stimulus intensity required to evoke a response of 50μV in at least 5 out of 10 successive trials.[38]

MEP latency and maximal MEP amplitude

Following hotspot and RMT determination, participants were instructed to perform a continuous abduction of the index finger at 10% maximal voluntary contraction (MVC). MVC was determined as the highest plateau of force production from index finger abduction over two trials, using a custom-built force transducer. During the 10% MVC, participants received 11 TMS stimulations over the contralateral hemisphere at 140% RMT. Participants could monitor their force production during the stimulations and were told to ignore perturbations elicited by the TMS stimulations.

Peripheral nerve stimulation

Peripheral nerve stimulation was applied to the ulnar nerve at the wrist point using a constant current generator (Digitimer, Cambridge Electronic Design, Cambridge, UK) in order to determine the maximal M-wave and the F-wave latency. 200 μs pulses were applied using a bipolar surface stimulation electrode. The stimulation intensity was gradually increased until no further increase in the peak-to-peak amplitude could be observed. Following M_{max} determination, we applied 10 electrical stimulations at M_{max} intensity for F-wave latency assessment. Data was saved on an internal server for off-line processing.

Data analysis

Electrophysiology

SEPs

Individual sweeps were filtered with a second order butterworth filter with a low cut of 0.1 Hz and a high cut of 250 Hz. The N20 latency was determined as the latency of the peak of the first deflection in the average trace of 500 stimulations applied to the wrist. N20 latency was determined manually for each hemisphere.

MEP_{max}

Participants force production was closely monitored during stimulations and trials were discarded if the required force level was not met. Participants were blinded to the force production during the first trial, which was always discarded. From the remaining trials the peak to peak amplitude of the recorded MEP was calculated and the maximal amplitude was chosen as the MEP_{max}. MEP_{max} amplitudes were normalized to individual M_{max} amplitudes.

Central motor conduction time (CMCT)

The CMCT was calculated as proposed by,[37] by subtracting the peripheral motor latency from the cortical motor latency. Here, the cortical motor latency was determined as the first local minima observed before a deflection of the EMG signal more than the pre-stimulus maximum contraction level plus 3 standard deviations of the mean rectified pre-stimulus contraction level. This observation was based on the mean of all remaining MEP_{max} recordings. The peripheral motor latency was determined as:

$$PML = 0.5 \times (F + M - 1) \qquad (1)$$

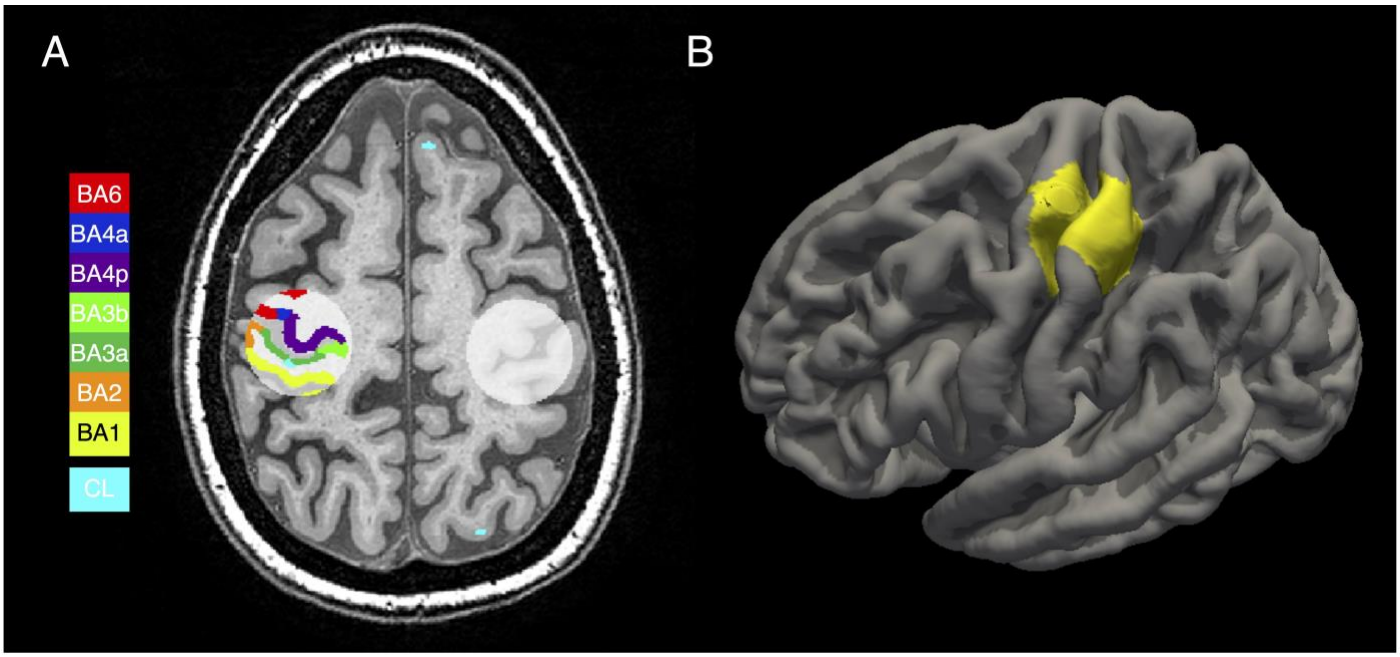
With F being the fastest observed F-wave latency and M as the fastest M-wave latency measured as the first local minima before a 2 SD deviation in the EMG from the rectified background noise level was observed.

Data transformations

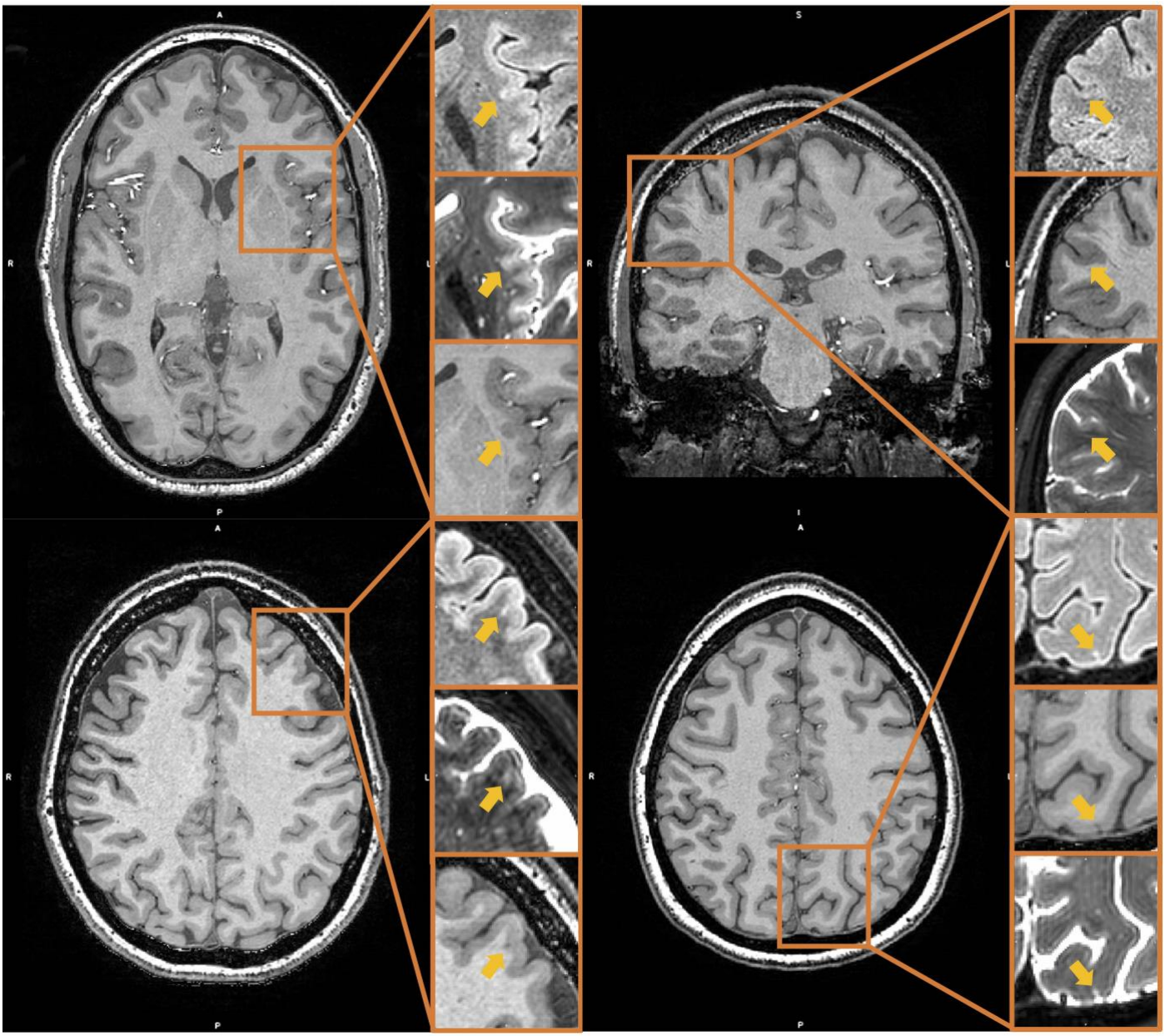
Due to a non-normal distribution of residuals from the liner mixed model with CMCT as the outcome variable, CMCT was log-transformed. This approach did not solve the similar issue

for the N20 latency. Therefore, the N20 latency was Box-Cox transformed with an optimal lambda, calculated using the *car* package in *RStudio*.

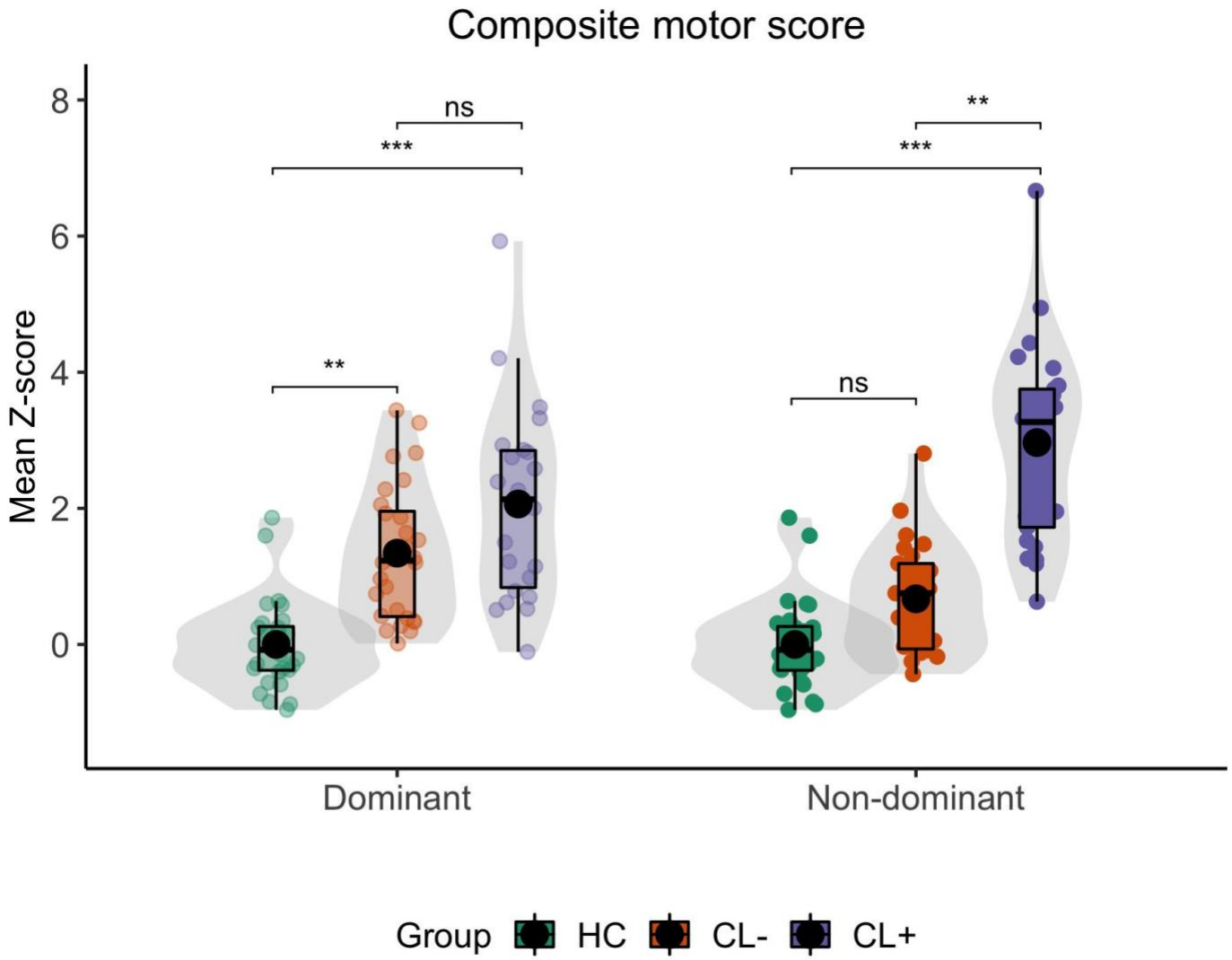
Supplementary figures



Supplementary figure 1. Detailed depictions of the hand knob region of interest. A) Single-slice depiction of the hand-knob region of interest including Broadmann area segmentations from *freesurfer*. **B)** Median handknob region of interest projected onto the FS average surface form fressurfer. Abbreviasions: BA = Broadmann area, CL = cortical lesion



Supplementary figure 2. Exemplary cortical abnormalities in the healthy control cohort. Depictions of four out of the seven cortical abnormalities detected in our HC cohort with enlargements of the FLAIR, MPRAGE and T2w sequences (top to bottom).



Supplementary figure 3. Predicted values from the interaction model. Box- and violin plots of predicted values from the mixed linear model including an interaction term for group*hand-dominance for healthy controls and patients with (CL+) and without cortical lesions (CL-) in the contralateral hand knob region of interest. Boxplots include median and interquartile range as horizontal lines and the mean as a black dot. Whiskers indicate the 5th and 95th percentile. Individual predicted data points are plotted on either side of the boxplot with low opacity representing the dominant hand and high opacity the non-dominant hand. *P<0.05, **P<0.01, *** P<0.001, ns = not significant.