Corticospinal tract integrity is related to primary motor cortex thinning in relapsing–remitting multiple sclerosis

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M.M. Lagana and E. Tavazzi contributed equally to the paper.

Abstract

Background: The relationship between white matter injury and cortical atrophy development in relapsing–remitting multiple sclerosis (RRMS) remains unclear.

Objectives: To investigate the associations between corticospinal tract integrity and cortical morphology measures of the primary motor cortex in RRMS patients and healthy controls.

Methods: 51 RRMS patients and 30 healthy controls underwent MRI examination for cortical reconstruction and assessment of corticospinal tract integrity. Partial correlation and multiple linear regression analyses were used to investigate the associations of focal and normal appearing white matter (NAWM) injury of the corticospinal tract with thickness and surface area measures of the primary motor cortex. Relationships between MRI measures and clinical disability as assessed by the Expanded Disability Status Scale and disease duration were also investigated.

Results: In patients only, decreased cortical thickness was related to increased corticospinal tract NAWM mean, axial and radial diffusivities in addition to corticospinal tract lesion volume. The final multiple linear regression model for PMC thickness retained only NAWM axial diffusivity as a significant predictor (adjusted $R^2=0.270$, $p=0.001$). Clinical measures were associated with NAWM corticospinal tract integrity measures.

Conclusions: Primary motor cortex thinning in RRMS is related to alterations in connected white matter and is best explained by decreased NAWM integrity.

Keywords: Multiple sclerosis, MRI, brain atrophy, diffusion tensor imaging, cortical atrophy, corticospinal tract

Introduction

Multiple sclerosis (MS) is a chronic inflammatory disease affecting the central nervous system which is now accepted to have a neurodegenerative component as well. While white matter injury was historically the primary research focus, the role of grey matter pathology has increasingly come under closer scrutiny. Cortical atrophy, primarily as evidenced by thinning of the cortical ribbon, is a widely recognized feature of MS and has been shown to occur early in the disease. Moreover, disability progression and cognitive impairment appear to be heavily dependent on irreversible grey matter loss.

The exact mechanisms that contribute to cortical atrophy in MS remain to be fully elucidated. There is clear evidence, though, of direct injury within the cortex, as evidenced by demyelination, neuronal loss, activated microglia and neurite transection. However, focal lesions within the white matter itself, as well as injury within the so-called normal appearing white matter (NAWM), are also likely to play a role via mechanisms such as retrograde myelinoaxonal degradation.

While several studies have investigated the global impact of white matter-induced grey matter injury using different MRI techniques, only a limited...
number\textsuperscript{1,11,12} have assessed the relationship between functionally and anatomically connected areas. Diffusion tensor imaging (DTI) is a technique which allows for both the in vivo reconstruction of white matter tracts\textsuperscript{13} and the assessment of tissue integrity changes in the NAWM.\textsuperscript{10} Meanwhile, advanced cortical reconstruction methodologies have been developed for regional parcellation and measurement of cortical morphology. Against this background, we aimed to investigate the relationship between cortical changes in a specific region along with measures of white matter integrity in a functionally and anatomically connected tract. As motor dysfunction is primarily responsible for irreversible disability in MS, we chose the primary motor cortex (PMC) and the corticospinal tract (CST), respectively. We hypothesized that altered white matter integrity as measured in both focal lesions and NAWM within the CST would be related to cortical morphology.

**Methods**

**Subjects**

This study included 51 patients with relapsing–remitting MS (RRMS). At the time of MRI acquisition, MS patients were relapse- and steroid-free within the last three months. RRMS patients with any other pre-existing medical condition were also excluded. Clinical disability in patients was quantified via the Expanded Disability Status Scale (EDSS). Thirty healthy controls were recruited from volunteers who had a normal neurological examination with no history of neurological, psychiatric, cardiovascular or metabolic disorders. All study participants provided written informed consent. The study was approved by the ‘Don Carlo Gnocchi Foundation’ ethics committee, Milan, Italy.

**MRI acquisition**

All scans were acquired on the same 1.5T MRI scanner (Siemens Magnetom Avanto, Erlangen, Germany) with a 12-channel head matrix coil. The following sequences were acquired: 1) dual-echo turbo spin echo proton density (PD)/T2-weighted (repetition time (TR)= 2650ms; echo time (TE)= 28/113ms; echo train length= 5; 50 contiguous 2.5-mm thick axial slices; 1mm\textsuperscript{2} in-plane resolution); 2) three-dimensional T1-weighted magnetization-prepared rapid gradient echo (MP-RAGE) (TR= 1900ms; TE= 3.37ms; inversion time (TI)= 1100ms; flip angle= 15; 176 contiguous, 1-mm thick axial slices; 1mm\textsuperscript{2} in-plane resolution); 3) diffusion weighted (DW) pulsed-gradient spin echo planar (TR= 7000ms; TE= 94ms; 50 contiguous, 2.5-mm thick axial slices; 2mm\textsuperscript{2} in-plane resolution; diffusion gradients applied in 12 non-collinear directions with a b-value= 900 s/mm\textsuperscript{2}; number of runs = 2). All sequences were acquired with full coverage of the brain and slices parallel to the subcallosal plane.

**Lesion segmentation.** White matter lesions were segmented by an experienced neurologist on the PD-weighted scans (Figure 1) with JIM software (http://www.xinapse.com/) version 5, which utilizes a semi-automated, local thresholding technique. The corresponding T2-weighted scan was used to increase confidence in lesion identification. To avoid the impact of T1 hypointensities on tissue segmentation, 3D T1 images were preprocessed using the lesion_filling tool.

**Morphological reconstruction.** Cortical reconstruction was performed on the 3D T1 images (lesion filled, in the case of MS patients) using the FreeSurfer package\textsuperscript{15,16} (http://www.freesurfer.net/) version 5.3 (Figure 2). Quality control was performed at all steps of the pipeline and manual corrections were made as necessary. The DKT40 atlas\textsuperscript{17} was then used to extract thickness and surface area measures for the precentral gyrus (corresponding to the PMC) and transverse temporal gyri (corresponding to the primary auditory cortex (PAC)); the latter was used as a control region.
Thalamic and total intracranial volumes (TIVs) were also obtained.

**CST measures.** DW images were corrected for eddy current and patient motion induced distortions using the eddy_correct tool. Then, the tensor was calculated for each voxel using dtifit, from which voxel-wise maps of fractional anisotropy (FA), mean diffusivity (MD), axial diffusivity (AD) and radial diffusivity (RD) were obtained. As white matter lesions may hinder conventional tractography, we used a previously generated in-house probabilistic CST atlas, derived from a completely separate sample of healthy controls. Briefly, the CST of 25 healthy controls was reconstructed with deterministic tractography by placing inclusion masks in the precentral gyrus and the anterior half of the posterior limb of the internal capsule. The reconstructed tracts were then warped into standard Montreal Neurological Institute (MNI) space and averaged such that voxel intensities represent the probability of corresponding to the CST.

Using FNIRT, subject-specific FA images were warped to the FMRIB_58_FA image. Trilinear interpolation was used and lesions were excluded as part of the cost function. The calculated warp field was then used to create MD, AD, RD and binarized lesion maps. Warped lesion maps were thresholded at 0.5 and re-binarized. The CST atlas was then used to extract the aforementioned DTI-based parameters for all subjects (Figure 3). In the MS patients, we calculated measures within both the NAWM and lesions. CST measures were computed by weighting the DTI indexes by the probability of a given voxel corresponding to the CST in the atlas. Finally, we computed weighted lesion volumes within the CST in a similar manner as for the DTI parameters.
To provide confirmation of the atlas-based tractography results, we used the Tract Based Spatial Statistics (TBSS) pipeline. Rather than performing a full voxel-wise analysis, we extracted mean DTI parameters within the CST portion of the TBSS skeleton. Lesion probability maps. Lesion probability maps (LPMs) within the CST were created as previously described. Briefly, individual lesion masks were all put into a common space and then averaged. The resulting map corresponds to the probability of a given voxel corresponding to a lesion. Permutation based inference testing was then used to test for voxel-wise associations between the spatial positions of CST lesions and cortical thickness measures, with age and sex included as nuisance covariates. The probabilistic CST atlas was binarized for use as a mask to restrict the LPM analyses only to lesions within the CST. Voxels with \( p < 0.05 \) corrected for family-wise error rate were considered significant.

Statistical analysis. Statistical analyses were performed using SPSS (version 21; IBM Corp., Armonk, NY, USA). Differences in demographic characteristics between the groups were assessed using Student’s \( t \)-test and Fisher’s exact test, as appropriate. Normality of the distribution of variables was assessed using the Kolmogorov–Smirnov method. T2 lesion volumes were logarithmically transformed due to positive skew. Although measures for the left and right hemispheres were obtained separately, total or average values, as appropriate, were calculated to reduce the number of comparisons.

Group differences between CST measures were assessed using an ANCOVA model, controlling for age and sex. For surface area measures, \( \frac{2}{3} \sqrt[3]{TIV} \) was also included as a covariate. The same covariates were included when examining partial correlations between cortical, CST and clinical outcomes. Next, multiple linear regression (MLR) analyses were used to assess the contribution of variables showing univariate correlations with cortical measures and clinical measures. Covariates were input into first block (enter method) and MRI measures in the second (forward-stepwise method) with cortical and clinical outcome measures as the dependent variables. We also assessed for interactions between group and NAWM CST indices when predicting cortical measures. As the disease duration and EDSS distributions were positively skewed, we repeated all analyses using the following sub-groups to exclude potential effects due to outliers: 1) disease duration<20 years; 2) EDSS< 6; 3) disease duration<20 years and EDSS<6. \( p \)-values <0.05 were considered significant using two-tailed tests.

Results

Demographic, clinical and MRI characteristics
Thirty healthy controls and 51 RRMS patients were enrolled in this study. Table 1 provides an overview of the demographic and clinical characteristics. No significant demographic differences between the two groups were found. Table 2 shows the imaging characteristics of the groups. No healthy controls presented with T2 lesions. RRMS presented with a mean global T2 lesion volume of 12.23 ± 13.37ml (median 6.94ml, range 0.79–61.16ml). For CST DTI indices in the NAWM, RRMS patients presented with significantly lower FA, higher MD and RD. RRMS patients presented with significantly decreased PMC thickness.

Relationships with cortical measures in healthy controls and patients
Partial correlations between cortical measures and other MRI parameters are shown in Table 3.

In the controls group, PMC surface area, but not cortical thickness, correlated with MD and AD within the
CST. With MLR, the final model retained as significant predictors: TIV ($\beta = 0.689$, $p=0.001$), age ($\beta = -0.460$, $p=0.004$) and AD ($\beta = -0.355$, $p=0.021$) ($F(3,20)=11.789$, $p<0.001$, adjusted $R^2 =0.652$).

In the RRMS group, PMC thickness, but not surface area, correlated with all three CST diffusivity measures and with the volume of CST lesions. With MLR, the final model retained as a significant predictor only NAWM AD ($\beta = -0.426$, $p=0.001$) ($F(3,47)=7.167$, $p<0.001$, adjusted (adj.) $R^2=0.652$).

When investigating the entire cohort, the interaction term between group and NAWM CST AD was significant ($p=0.028$) in predicting PMC thickness. For PMC surface area, significant interaction terms between group and NAWM CST MD ($p=0.025$) and RD ($p=0.026$) were found.

Thalamic volume was not associated with cortical outcomes. PAC measures were not related to any other MRI measures.

Relationships between clinical and MRI measures in patients
Partial correlations with clinical outcomes are shown in Table 4. Both disease duration and EDSS correlated with measures of CST NAWM FA and all three diffusivity measures. In addition, EDSS, but not disease duration, correlated with both increased CST lesion volume and decreased PMC thickness. With MLR, the final model for EDSS retained as significant predictors: age ($\beta = 0.302$, $p=0.008$), CST lesion volume ($\beta = -0.458$, $p<0.0001$) and PMC thickness ($\beta = -0.254$, $p=0.033$) ($F(4,46)=13.007$, $p<0.001$, adj. $R^2=0.490$).

Table 2. Corticospinal tract and cortical measures in healthy controls and relapsing–remitting multiple sclerosis.

<table>
<thead>
<tr>
<th></th>
<th>HC ($N=30$)</th>
<th>RRMS ($N=51$)</th>
<th>$p$</th>
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<tr>
<td><strong>CST measures</strong></td>
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<td>NAWM</td>
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<tr>
<td>FA ATLAS</td>
<td>0.50 (0.02)</td>
<td>0.47 (0.02)</td>
<td>$&lt; 0.0001$</td>
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<tr>
<td>FA TBSS</td>
<td>0.54 (0.02)</td>
<td>0.51 (0.02)</td>
<td>$&lt; 0.0001$</td>
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<tr>
<td>MD ATLAS</td>
<td>0.77 (0.02)</td>
<td>0.81 (0.05)</td>
<td>$&lt; 0.0001$</td>
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<tr>
<td>MD TBSS</td>
<td>0.73 (0.01)</td>
<td>0.76 (0.03)</td>
<td>$&lt; 0.0001$</td>
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<tr>
<td>AD ATLAS</td>
<td>1.22 (0.02)</td>
<td>1.25 (0.05)</td>
<td>0.013</td>
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<tr>
<td>AD TBSS</td>
<td>1.23 (0.02)</td>
<td>1.23 (0.03)</td>
<td>0.885</td>
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<tr>
<td>RD ATLAS</td>
<td>0.54 (0.02)</td>
<td>0.60 (0.05)</td>
<td>$&lt; 0.0001$</td>
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<td>RD TBSS</td>
<td>0.49 (0.02)</td>
<td>0.52 (0.04)</td>
<td>$&lt; 0.0001$</td>
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<td><strong>T2 lesion</strong></td>
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<tr>
<td>Volume</td>
<td>–</td>
<td>0.49 (0.80)</td>
<td>0.20 (0.01–3.2)</td>
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<tr>
<td>FA ATLAS</td>
<td>–</td>
<td>0.37 (0.07)</td>
<td>–</td>
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<tr>
<td>MD ATLAS</td>
<td>–</td>
<td>0.98 (0.15)</td>
<td>–</td>
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<tr>
<td>AD ATLAS</td>
<td>–</td>
<td>1.37 (0.16)</td>
<td>–</td>
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<tr>
<td>RD ATLAS</td>
<td>–</td>
<td>0.79 (0.16)</td>
<td>–</td>
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<tr>
<td><strong>Cortical measures</strong></td>
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<tr>
<td>Thickness</td>
<td></td>
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<tr>
<td>Primary motor cortex</td>
<td>2.39 (0.14)</td>
<td>2.23 (0.26)</td>
<td>0.017</td>
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<tr>
<td>Primary auditory cortex</td>
<td>2.41 (0.27)</td>
<td>2.32 (0.28)</td>
<td>0.305</td>
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<tr>
<td><strong>Surface area</strong></td>
<td></td>
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<tr>
<td>Primary motor cortex</td>
<td>9647 (983)</td>
<td>9576 (1071)</td>
<td>0.998</td>
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<tr>
<td>Primary auditory cortex</td>
<td>781 (132)</td>
<td>731 (109)</td>
<td>0.075</td>
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</table>

HC: healthy controls; RRMS: relapsing–remitting multiple sclerosis; N: number; CST: corticospinal tract; NAWM: normal appearing white matter; FA: fractional anisotropy; MD: mean diffusivity; AD: axial diffusivity; RD: radial diffusivity; TBSS: tract based spatial statistics;

Data are presented as mean (standard deviation) except for CST T2 volume where the median (range) is also provided. FA is a dimensionless index. MD, AD and RD are given in mm$^2$/s * 10$^{-3}$. Lesion volume is given in millilitres. Thickness measures are given in mm whereas surface area measures are in mm$^2$. $p$ values were calculated using ANCOVA, controlling for age and sex; the $\sqrt{\text{TIV}}$ was included as well for surface area comparisons. Significant differences are shown in bold.
Table 3. Partial correlations with cortical measures in healthy controls and relapsing–remitting multiple sclerosis.

<table>
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<tr>
<th></th>
<th>HC</th>
<th>RRMS</th>
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<th>HC</th>
<th>RRMS</th>
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<th>HC</th>
<th>RRMS</th>
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<tr>
<td></td>
<td>Thickness</td>
<td>Surface area</td>
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<td>Surface area</td>
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<tr>
<td>Thalamic volume</td>
<td>-0.156 (0.488)</td>
<td>-0.150 (0.515)</td>
<td>0.140 (0.533)</td>
<td>0.007 (0.975)</td>
<td>0.276 (0.055)</td>
<td>0.067 (0.653)</td>
<td>0.165 (0.258)</td>
<td>0.085 (0.564)</td>
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<td>NAWM CST measures</td>
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<tr>
<td>FA</td>
<td>0.072 (0.749)</td>
<td>0.236 (0.304)</td>
<td>0.190 (0.397)</td>
<td>0.346 (0.125)</td>
<td>0.264 (0.067)</td>
<td>-0.064 (0.667)</td>
<td>0.084 (0.566)</td>
<td>0.062 (0.677)</td>
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<tr>
<td>MD</td>
<td>0.219 (0.328)</td>
<td>-0.476 (0.029)</td>
<td>-0.065 (0.773)</td>
<td>-0.313 (0.167)</td>
<td>-0.440 (0.002)</td>
<td>0.038 (0.797)</td>
<td>-0.104 (0.476)</td>
<td>-0.154 (0.295)</td>
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<tr>
<td>AD</td>
<td>0.411 (0.057)</td>
<td>-0.500 (0.021)</td>
<td>0.128 (0.571)</td>
<td>-0.131 (0.570)</td>
<td>-0.448 (0.001)</td>
<td>0.025 (0.867)</td>
<td>-0.104 (0.479)</td>
<td>-0.174 (0.237)</td>
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<tr>
<td>RD</td>
<td>0.106 (0.638)</td>
<td>-0.372 (0.097)</td>
<td>-0.114 (0.614)</td>
<td>-0.332 (0.142)</td>
<td>-0.426 (0.002)</td>
<td>0.048 (0.746)</td>
<td>-0.104 (0.479)</td>
<td>-0.139 (0.347)</td>
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<td>Lesional CST measures</td>
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<tr>
<td>FA</td>
<td>-</td>
<td>-</td>
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<td>-</td>
<td>0.148 (0.317)</td>
<td>-0.051 (0.734)</td>
<td>-0.086 (0.560)</td>
<td>0.077 (0.605)</td>
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<tr>
<td>MD</td>
<td>-</td>
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<td>-</td>
<td>-</td>
<td>-0.190 (0.196)</td>
<td>0.004 (0.979)</td>
<td>-0.026 (0.858)</td>
<td>-0.287 (0.050)</td>
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<tr>
<td>AD</td>
<td>-</td>
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<td>-</td>
<td>-0.150 (0.308)</td>
<td>-0.007 (0.964)</td>
<td>-0.086 (0.560)</td>
<td>-0.276 (0.061)</td>
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<tr>
<td>RD</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.191 (0.194)</td>
<td>0.010 (0.949)</td>
<td>0.008 (0.959)</td>
<td>-0.264 (0.073)</td>
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<tr>
<td>Volume</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.327 (0.022)</td>
<td>-0.039 (0.791)</td>
<td>-0.173 (0.234)</td>
<td>-0.265 (0.482)</td>
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</table>

Partial correlations were calculated controlling for age and sex; the $\sqrt{\frac{V}{TIV}}$ was included as well for surface area comparisons. Data shown as $r$ (p). Significant correlations are shown in bold. HC: healthy controls; RRMS: relapsing–remitting multiple sclerosis; CST: corticospinal tract; NAWM: normal appearing white matter; FA: fractional anisotropy; MD: mean diffusivity; AD: axial diffusivity; RD: radial diffusivity.
PAC thickness and surface area were not related to either of the clinical measures.

**Lesion probability mapping**

Within the CST, lesion probability was highest (43.1%) in the left CST (MNI standard coordinate: x=−20, y=−24, z=28). PMC thickness correlated with lesion probability throughout much of the CST (Figure 4 and Table 5). No significant voxels were found for the PAC.

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**TBSS results**

With the exception of NAWM AD, group differences were of a similar magnitude between results...
Sub-group analyses

Sub-group analyses with disease duration <20 years retained 39 patients; EDSS <6 retained 36 patients; disease duration <20 years and EDSS <6 retained 33 patients. Results obtained with the reduced cohorts were in line with those of the full cohort (data not shown).

Discussion

In this cross-sectional study of patients with RRMS, we showed that MRI measures of white matter integrity are related to cortical thickness of an anatomically and functionally connected cortical area. Although the link between grey matter and white matter damage in MS has been the subject of intense study, most reports have examined either global relationships or only connected white matter–grey matter regions. Thus, we included the PAC as a control region since it has no connections with the CST. While stronger relationships between CST integrity and morphological measures of the PMC are more plausible than with the PAC, the complete lack of any correlations with the latter may be somewhat surprising considering the widespread damage caused by MS throughout the brain, including within the auditory cortex. One might expect a relationship between anatomically distinct areas merely due to overall disease burden. In this context, our results underline the fact that regional investigations are likely to be more informative when studying the interplay between grey matter and white matter pathology.

We found robust relationships between PMC and CST integrity measures in RRMS patients. While no DTI metrics within the CST lesions were associated with PMC thickness, all of them (i.e. FA, MD, RD, AD) were significantly correlated with it in the NAWM. This is in line with whole brain associations recently reported by Steenwijk et al. The finding of very similar correlation coefficients may suggest that the degree of tract-specific integrity and its relationship to thinning in connected cortical areas may be similar throughout the brain. On the other hand, the rate of cortical thinning should scale with the relative degree of connectivity for a given area if it is in part due to axonal transection following focal white matter injury. Thus, the investigation of other tracts and corresponding cortical regions may help elucidate this question. It should be noted though that while lesion volume and location was related to PMC thickness in both univariate correlation and LPM analysis, NAWM AD was the only MRI variable retained in the regression analysis. Thus, NAWM, rather than focal measures, may capture more of the overall tissue injury. Finally, we did not find any associations between CST integrity and PMC surface area, contrary to previously published data. However, while cortical thinning has been widely described in MS, differences in surface area have not been consistently reported.

Although FA was significantly reduced in the NAWM of the CST in RRMS patients compared with healthy controls and in lesions with respect to NAWM, it was not related to PMC measures. Few studies have investigated the relationship between white matter tract FA and cortical thickness, with conflicting findings, likely due to patient cohort differences. Indeed, the longer disease duration reported in other studies might have led to increased tissue microstructure damage. This lends support to the notion that FA may be less sensitive than diffusion coefficient indices to the amount of underlying tissue damage. In addition, one should keep in mind that FA will remain relatively stable in the event that there is no preferential change in AD or RD. Finally, one longitudinal study reported that DTI indices in the CST did not change in a consistent pattern in MS patients. Thus, additional longitudinal studies are needed to better understand the evolution of diffusion coefficient and FA changes in both lesions and the NAWM and their relation to cortical thinning.
Furthermore, increased EDSS was related to PMC thinning to a similar degree as has been previously reported. Such a relationship might be expected considering that EDSS is heavily weighted towards motor dysfunction. The fact that the FreeSurfer PMC parcellation is larger than the area responsible for motor/ lower limb function may explain why the relationship is not stronger. Thus, the application of other clinical scales, including those able to capture upper limb dysfunction, may indeed improve the relationships we found between MS disability and cortical damage.

In the healthy control sample, we unexpectedly found correlations between MD and AD in the CST and PMC surface area, but not thickness. Age was included as a covariate as it has been shown to correlate with both increased white matter diffusivity and decreased cortical surface area and thickness. Sex was also included due to expected differences. A residual nonlinear effect of sex after covariation can be excluded as these relationships were still evident when investigating males and females separately. Meanwhile, no such relationships were found in the MS sample despite not differing in PMC surface area. It is important to consider that several of the interaction terms between group and diffusivity indices were significant when predicting cortical measures in the cohort as a whole. This suggests that the relationship between measures of white matter and connected grey matter change as a result of the disease process. These findings warrant further investigation in a larger sample to better understand the underlying dynamics.

There are a number of limitations in this study that need to be considered. As with any cross-sectional study, it is not possible to draw any firm conclusions regarding the temporal evolution between the investigated measures. It should also be noted that the FreeSurfer precentral gyrus parcellation is larger than the area to which the CST connects. Thus, damage in connecting tracts that we did not consider may also contribute to PMC injury. Moreover, we only considered a single white matter tract and associated cortical region. As such, longitudinal studies investigating a larger number of tracts/regions are warranted to better understand the interplay between white matter–grey matter injury. Additionally, the number of acquired diffusion directions in this study was quite small by current standards. These sequences were acquired during clinical examinations though, limiting the available amount of time. Regardless, all participants were scanned using the same protocol, minimizing any systematic bias in the results. Moreover, the CST atlas used in this study was generated from a separate healthy control sample and we subsequently confirmed our findings with TBSS. Consequently, despite the limited DTI protocol, the overall interpretability of the results should not be affected. It should also be noted that our study focused primarily on the relationship between grey matter atrophy and damage in the white matter. While we did not find clear evidence of thalamic atrophy being related to PMC thinning, cortical pathology is likely explained as well in part by primary causes such as neuronal/axonal damage within the cortex itself. We lacked a method to visualize cortical lesions and could not assess their contribution to cortical thinning. Finally, we did not distinguish between juxtacortical and ‘pure’ white matter lesions in our analysis, which may provide further insight in this regard.

In conclusion, the present study demonstrated that focal as well as diffuse tissue microstructure alterations in the CST are associated with thinning of the PMC in RRMS. This finding highlights the importance of studying anatomically and functionally related areas of the brain for a better understanding of the pathogenesis of the damage in MS. Serial MRI studies are needed to unravel the temporal–spatial dynamics of white matter injury and cortical atrophy.

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Conflict of interest
The authors declare that there is no conflict of interest.

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References
4. Nygaard GO, Walhovd KB, Sowa P, et al. Cortical thickness and surface area relate to specific symptoms in early relapsing–remitting multiple sclerosis. Mult...
Multiple Sclerosis Journal

Scler. Published online before print 19 August 2014. DOI: 10.1177/135245851453811.


