What does (quantitative) MRI of the MS cortical gray matter measure? A post mortem imaging exploration.

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Objective:
Cortical lesions are abundant in the cortical gray matter (CGM) of multiple sclerosis (MS) patients, but only a small percentage of these lesions can be visualized by conventional magnetic resonance imaging (MRI). Quantitative MRI techniques have been reported to be more sensitive to cortical damage, but the histopathological correlates of quantitative MRI changes in the MS CGM are unclear. The aim of this study was to define the underlying pathology of cortical quantitative MRI changes, and to compare their sensitivity to cortical pathology. Additionally, we investigated whether MRI-visible cortical lesions differ from MRI-invisible cortical lesions in terms of histopathology.

Materials and Methods:
16 formalin-fixed hemispheric brain slices from 10 patients with chronic MS were analyzed using quantitative and semiquantitative histopathology ratings of neuroaxonal density, astrogliosis, myelin, microglial reactivity, and blood-brain barrier leakage (*Figure2*), as well as qualitative (dual-echo T2 spin-echo (TR/TE/NEX: 2755ms/45ms/90ms/2; FoV: 80x128mm; matrix size: 160x256; slice thickness: 3mm)) and quantitative (T1-, T2- and MTR maps, *Figure1*) MRI at 1.5T. For T1-mapping, a Flash3D sequence with a range of flip angles (2-25°) and B1 correction was used, and a CPMG sequence for T2 mapping. For MTR, Flash3D images with a 7.68 ms Gaussian pre-pulse (frequency offset 1500 Hz, effective flip angle 500°) were used. Cortical lesions were scored on qualitative MRI and PLP-stained sections (*Figure2*). A region-of-interest (ROI) approach was applied to compare MRI-visible to MRI-invisible GM lesions and non- lesional (NL) GM (*Figure1*). Overall GM histogram parameters of the quantitative maps were correlated with histopathological measurements.

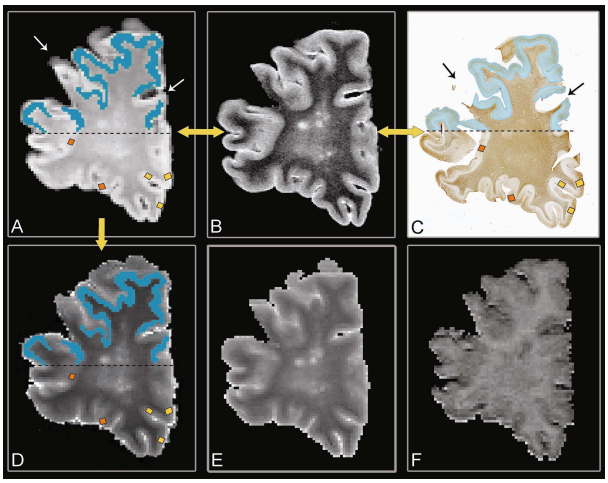
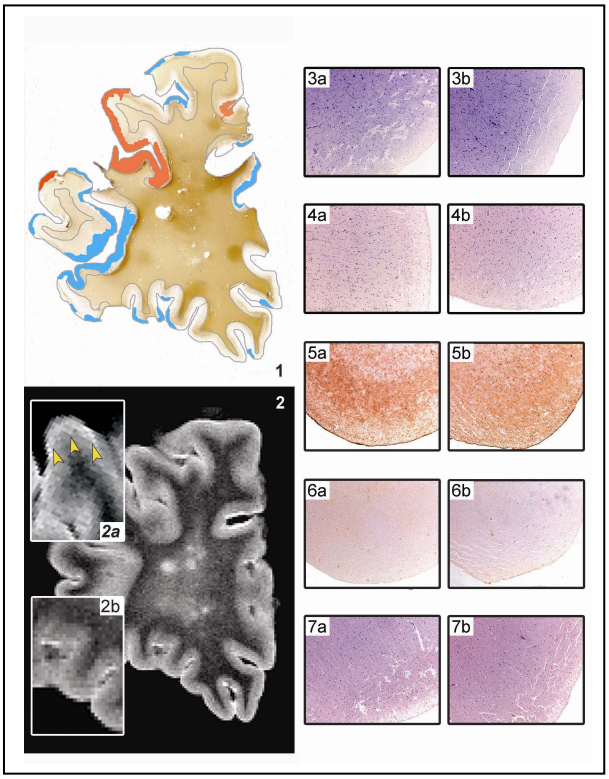


Figure 1. Demonstration of MRI-to histopathology matching and placement of regions of interest (ROIs) on a post-mortem multiple sclerosis brain slice. A: native image for T1-map, B: Proton-Density weighted image, C: Tissue section stained for glial fibrillary acidic protein (GFAP), D: T1 map, E: T2 map, F: magnetization-transfer map. Each of the tissue sections (C) were matched separately with the native images (A), using the Proton-density weighted image as reference (B). Arrows on image A and C indicate inconsistent areas which were carefully considered in the matching procedure. For global analysis of the gray matter, the whole cortex was outlined on native images and histological sections, and these ROIs were copied on the calculated maps (blue areas, demonstrated on the upper half of images A,C and D). Similarly, small ROIs were placed for focal analysis of the grey matter. (yellow: ROIs in the non-lesional grey matter, orange: ROIs in grey matter lesions; demonstrated on the lower half of images A,C, and D).



Results:
Cortical GM lesions (n =187) displayed a loss of neuropil (cell bodies, axons, dendrites) as compared to NLGM (4.2% loss of staining intensity in Bodian- and 12.3% loss in Nissl-stained sections), as well as higher T1 and T2 measures (349.9±22.4 ms versus 323.7 ± 22.0 ms for T1 measures and 77.1 ± 2.7 ms versus 72.2 ± 2.6 ms for T2 measures, P<0.05). Cortical GM lesions that were visible on conventional MRI did not differ from MRI-invisible GM lesions in terms of underlying histopathology or in terms of quantitative MRI measures (*Figure2*). However, MRI-visible lesions were found to be significantly larger than their invisible counterparts (mean 13.3 ± 1.7mm² versus 6.9 ± 1.3mm²; *P*=.001). GM lesion size correlated with the overall percentage of demyelination in the cortex of MS patients (r=0.78, *P*<0.01). Although the overall percentage of GM demyelination per MS slice was generally low in the brain slices, demyelination still significantly correlated with reduced mean MTR and MTR histogram peak position.

Conclusion
Quantitative MRI measurements are highly sensitive to cortical demyelination, and are more likely to reflect the extent of cortical demyelination than any significant pathology in the NL GM. Conspicuity of cortical GM lesions on conventional MRI was determined by lesional size, rather than by any distinctive histopathological differences between lesions, and the presence of lesions with greater size was associated with a higher overall cortical lesion load.

Figure 2. Comparison between MRI- visible and invisible cortical lesions. 1: Cortical lesions were assessed on proteolipid- protein stained tissue sections (original magnification x0.7), and after comparison with the corresponding MRI images, marked as visible (red) and invisible (blue). 2: Corresponding proton density weighted MR image of the same brainslice. 2a: MRI-visible lesion. Note the subtle signal intensity increase that could be deteced after direct comparison with the proteolipid stained tissue section. 2b: MRI invisible lesion. 3a-7b: MRI visible lesions did not differ from MRI- invisible lesions in terms of histopathology. Left column, 3a-7a: histological sections of the MRI-visible lesion shown in 2a. Right column, 3b-7b: corresponding MRI-invisible lesion of figure 2b. Sections stained for neuropil (3a,b: Nissl stain, 7a,b: Bodian silver, both x50), antigen presenting cells (4a,b: HLA-DR, x50), astroglia (5a,b: glial fibrillary acidic protein stain, x25; though glial density seems to be lower in 5a as compared to 5b, no overall significant difference was detected for glial densities between MRI visible and invisible lesions), blood brain barrier leakage (6a,b: Fibrinogen, x25).