Sulcal and gyral crown cortical grey matter involvement in multiple sclerosis: a magnetisation transfer ratio study

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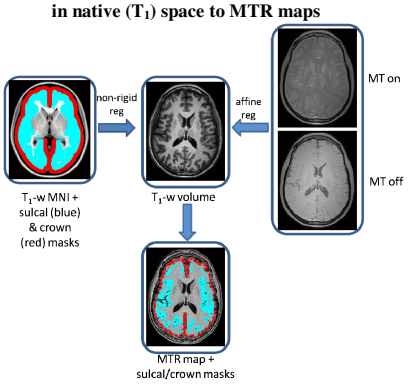
Target audience: Clinicians interested in grey matter pathology and its relationship with clinical status in multiple sclerosis (MS)

Purpose: To look for differences in MS disease effects on sulci and gyral crowns using magnetization transfer ratio (MTR) measurements in healthy volunteers and people with different MS clinical subtypes, and investigate associations with clinical features.

Introduction: Cortical grey matter (CGM) lesions are common in MS, and those detected with MRI are associated with physical and cognitive impairment (1). In *post mortem* studies, cortical demyelination appears to be mainly located in the sulci, particularly in SPMS (2-3), and it has been suggested that meningeal follicle-like B-cell aggregates may be linked with this. Pathological studies tend to obtain material from patients with long duration progressive MS, however there is evidence that cortical demyelination differs between subtypes of MS (4). It would therefore be useful to establish if the prevalence of cortical lesions and other abnormalities in sulci, as suggested by some histopathological studies, holds true *in vivo* in all MS subtypes. CGM lesions are much more difficult to identify using conventional MRI than white matter (WM) lesions (5-6), however an alternative way to indirectly assess them is to use a quantifiable MRI measure that is sensitive to pathology seen in cortical lesions. Magnetisation transfer ratio (MTR) measurements provide a quantitative measure of macromolecular tissue content, and WM MTR has been shown to correlate with myelin content and axonal density (7). MTR reductions can therefore also be anticipated in the cortex, particularly in cortical lesions (8).

The objective of this study was to develop a method to assess cortical MTR in sulci and gyral crowns in healthy controls (HC), and patients with relapsing-remitting (RR), primary and secondary progressive (PP and SP) MS, and determine if the differences suggested by histopathological studies are also detectable *in vivo*.

Figure 1: Flow chart illustrating registration processes to apply sulcal/gyral crown masks in native (T_1) space to MTR maps



Methods: ***Subjects:*** Thirty-one RR (mean age 43.9 years, median Expanded Disability Status Scale (EDSS) score 2.5), 14 SP (51.3 years, EDSS 6.5) and 16 PP (53.6 years, EDSS 6.0) MS patients and 32 healthy control (HC) subjects (mean age 37.0 years) were included in this study. Patients were clinically assessed, and their EDSS score and components of the MS Functional Composite (MSFC) (nine hole peg test [9HPT], 25 foot timed walk test [TWT] and Paced Auditory Serial Addition Test [PASAT], expressed as z-scores) were determined, the MSFC calculated using published normative data (9).

MR acquisition: Scanning was performed using a 3T Philips Achieva system (Philips Healthcare, Best, The Netherlands) with a 32-channel head coil and multi-transmit technology, using the following sequences (both acquired sagittally, with field-of-view (FOV) 256x256x180mm^3; voxel size1x1x1mm^3): i) T_1-weighted (T_1w) volumes using a 3D inversion-prepared (TI=824ms) gradient echo (FFE) sequence (TR/TE=6.9/3.1ms); flip angle=8°; ii) MTR data with a 3D slab selective spoiled gradient echo (FFE) sequence with 2 echoes (TR=6.4ms, TE1/TE2=2.7/4.3ms, α=9°) with and without Sinc-Gaussian shaped MT saturating pulses of nominal flip angle=360º, offset frequency 1kHz, duration 16ms applied prior to the excitation pulse.

Image Analysis: The MNI T_1 template was segmented using SPM8 (www.fil.ion.ucl.ac.uk/spm). Using WM and grey matter (GM) masks (split into hemispheres) and FSL (www.fmrib.ox.ac.uk/fsl) editing tools cerebral sulcal and crown GM masks were generated in addition to a global CGM mask. The MNI T_1 template was co-registered non-linearly using NiftyReg (10-11) to each individual subject's (lesion filled (12)) T_1w volume scan, and registration parameters were applied to masks to transform them to native space. T_1w volumes were also segmented in native space using SPM8, then in-house software was used to determine maximum likelihood tissue probability maps from the segmented data, thresholded at 90% to reduce the possibility of partial volume errors. MTR data for each subject were affine registered to their T_1w volume, and masks were applied to calculated MTR maps to give CGM, sulcal or gyral crown MTR values. The processes for applying the sulcal/gyral crown masks to MTR maps are illustrated in Figure 1. All statistical analysis was performed using SPSS version 11.0 for Windows (SPSS, Inc., Chicago, IL, USA).

Results: Mean CGM, crown and sulcal MTR values for each subject group are given in Table 1, where *= p<0.05, **= p<0.01, ***= p<0.001 compared to the control MTR value, tested via one-way ANCOVA tests, with post-hoc paired comparisons, with adjustments made for age and volume. Compared with HC, global CGM, sulcal and gyral crown MTR values were reduced in all MS groups (PPMS, cortical p<0.001, sulcal p<0.001 and gyral crown p<0.01, SPMS all p<0.001, and RRMS all p<0.01). SPMS patients had lower CGM, sulcal and gyral crown (p<0.001) MTR values than RRMS patients, and lower CGM (p<0.01), sulcal (p<0.01) and gyral crown MTR (p<0.001) than PPMS patients. There were no significant differences between PPMS and RRMS patients. Significant correlations with clinical scores were seen only in RRMS and SPMS groups and are shown in Table 2.

Table 1: Global cortical, sulcal and gyral crown mean MTR values for all subject groups, with percentage MTR for patients compared to the mean control value in italics (%)

	Cortical GM MTR (pu)	Gyral Crown MTR (pu)	Sulcal MTR (pu)
Controls	32.5 (± 0.62)	32.7 (± 0.69)	32.4 (± 0.62)
PPMS	31.2 (± 1.22)***, *3.29*	31.6 (±1.07)*, *3.23*	31.4 (±1.02)**, *3.19*
SPMS	31.4 (± 1.02)**, *7.22*	30.2 (±1.06)***, *7.56*	30.2 (±1.32)***, *6.81*
RRMS	30.2 (± 1.14)***, *2.85*	31.8 (±1.03)**, *2.86*	31.6 (±1.06)**, *2.52*
All MS	31.6 (± 1.10)**, *3.97*	31.5 (±1.23)***, *2.82*	31.3 (±1.23)***, *2.37*

Discussion: In MS, MTR abnormalities are apparent in CGM, gyral crown and sulcal cortex, particularly in those with relapse-onset disease (RR and SP MS). Overall, our results suggest that pathology, as reflected by MTR values, is similar throughout the cortex in RR and PPMS patients, but that gyral crown CGM abnormalities become more prominent in SPMS. In contrast to previously reported *post mortem* studies of cortical demyelination (2, 13), we did not find more marked MTR changes in sulcal compared to gyral crown CGM. *Post mortem* studies tend to be biased towards end-stage disease or atypical early presentations, and may not be representative of the majority of people living with MS. Further, current histopathological techniques cannot practically be used to systematically assess the whole brain. The potential effects of GM/CSF and GM/WM partial volume should be further investigated. Also, the progressive cohorts were smaller than the RRMS group, and data from larger groups may reveal more subtle subtype specific disease effects than could be observed it the present study.

Conclusion: Cortical pathology, as reflected by overall cortical MTR, is present in all MS subtypes and most pronounced in SPMS. A preferential disease effect on sulcal cortical regions was not observed. Cortical MTR abnormalities appear to be more clinically relevant in relapse-onset rather than progressive-onset MS.

Table 2: Significant correlations of CGM, gyral crown or sulcal MTR with clinical status measures in RRMS and SPMS

	CGM MTR (pu)	Gyral Crown MTR (pu)	Sulcal MTR (pu)
SPMS	TWT[a] (r=-0.95, p<0.001)		TWT (r=-0.83, p<0.01)
RRMS	EDSS[b] (r=-0.41, p<0.05), 9HPT[c] (r=0.49, p<0.01), PASAT[d] (r=0.46, p<0.05)	EDSS (r=-0.37, p<0.05), 9HPT (r=0.44, p<0.05), PASAT (r=0.50, p<0.01)	EDSS (r=-0.39, p<0.05), 9HPT (r=0.44, p<0,05), PASAT (r=0.47, p<0.01)
Relapse-onset (RR & SPMS)	EDSS (r=-0.37, p<0.05), DD[e] (r=-0.46, p=0.001) 9HPT (r=0.44, p<0.05), PASAT (r=0.50, p<0.01)	EDSS (r=-0.60, p<0.001)	EDSS (r=-0.55, p<0.001)

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Acknowledgements: *The authors would like to thank the MS Society of Great Britain and Northern Ireland and the Department of Health's NIHR Biomedical Research Centres funding scheme for funding. We would also like to thank all the participants of this study.*