
**Synopsis**
Magnetization transfer ratio histograms are sensitive to the evolution of normal appearing tissue pathology in multiple sclerosis (MS) yet it is unclear whether change over time is detectable in clinically early MS. For this reason, we have studied 23 subjects with early relapsing remitting MS and minimal disability together with 17 healthy controls. At baseline normal appearing grey and white matter MTR was significantly reduced in MS subjects and MTR change was detectable in both tissues at 1 and 2 year follow up.

**Background**
Magnetization transfer imaging (MTI) is able to detect the pathological change in multiple sclerosis (MS) both within lesions and in normal appearing tissue. (1,2) For this reason MTI has been proposed as a possible surrogate marker for disease progression. Early change in the normal appearing grey and white matter may play an important role in the development of cumulative disability (4) and so interventions that reverse or arrest this change are of interest. However, in order for such therapies to be developed it is important to detect and monitor early normal appearing tissue change, ideally, prior to the onset of permanent disability. The aim of this study was to ascertain whether early normal appearing grey and white matter change is detectable with MTR in a cohort of minimally disabled early relapsing remitting MS subjects. Previously, a cohort with clinically isolated syndromes and MS have demonstrated MTR change over time (5) yet it is not known whether MTR is sensitive to normal appearing change in a cohort typical of those recruited into treatment trials (namely those with early RRMS).

**Methods**

23 subjects with clinical definite early relapsing remitting MS (6) (19 female and 4 male; mean age 37 years, range 27-55; disease duration at baseline 1.9 years, range 0.5-3.7; Median expanded disability status score 1, range 0-3) and 17 healthy controls (9 female and 8 male; mean age 34.5. range 27-52) were studied. All subjects were imaged at baseline with 22 MS subjects and 13 controls imaged at 1 year (range 11-14 months) and 17 MS subjects and 10 controls imaged at 2 years (range 23-28 months). Patients were not on disease modifying medication prior to entry into the study. EDSS was determined at baseline. The study was approved by the ethics committee of the National Hospital for Neurology and Neurosurgery and all subjects gave informed consent.

Imaging was performed on a 1.5 Tesla Signa (GE, USA). The 2D SE MTI sequence has been described previously (7) and was acquired in controls and MS subjects at all time points. Repetition time (TR) was 1500ms with echo time (TE) of 19/90 ms. 28 contiguous 5mm axial slices covering the whole brain were acquired both with and without presaturation. The saturating MT pulse was 64 ms in duration and 2kHz off resonance. Saturated and unsaturated images were interleaved resulting in inherently registered saturated and unsaturated data sets. MTR was calculated from the short echo images on a pixel by pixel basis according to [(Mo-Ms)/Mo] x 100 percent units (pu) where Mo and Ms represent the signal intensities with and without the saturation pulse respectively. The acquisition time was 20 minutes.

Lesions were contoured using Disqimage (Plummer, Dept of medical physics and bioengineering, UCL, UK) on the unsaturated proton density weighted images. Segmentation of the T2 weighted images into grey matter, white matter and CSF segments was achieved using SPM99 (Wellcome Department of Cognitive Neurology, UK). A whole brain mask (that which excludes CSF and other non brain parenchyma) was generated in SPM99 and then applied to the calculated MTR map. A maximum likelihood algorithm, utilizing the grey and white matter probability outputs from SPM99 was then used to segment the MTR whole brain map into white and grey matter segments. Lesions were nulled to produce normal appearing white matter (NAWM) and normal appearing grey matter (NAGM). Partial volume voxels were minimized with a 10 pu threshold and 2 successive erosions of white matter and a single erosion of the grey matter (2 erosions of grey matter resulted in the too much of the grey matter being eliminated). Normalized NAWM, NAGM and whole white matter histograms were generated with a bin width of 0.1pu and a smoothing window of +/- 3pu. The mean MTR was extracted and comparisons between MS subjects and controls were achieved using linear modeling (SPSS 10.0). Gender and diagnosis were included as fixed factors with age as a covariate. The Wilcoxon signed ranks test was used to compare paired follow up and baseline values.

**Results**

Mean baseline T2 load was 16.6ml (range 2.1-47.7ml). Baseline NAWM and NAGM MTR was significantly reduced in MS subjects when compared with controls. MS NAWM MTR: 37.8pu (Standard deviation (SD) 0.6pu), Control white matter: 38.3pu (SD 0.5pu), p = 0.003. MS NAGM MTR: 32.0pu (SD 0.7pu), Control grey matter 32.4pu (SD 0.5pu), p = 0.001. Significant results were obtained even when age and gender were included as factors in the model. There was a significant effect of gender on grey matter yet no effect of age on either tissue.

Analysis of the 17 MS subjects at 2 years revealed a significantly reduced NAWM and NAGM MTR when compared with baseline. MS NAWM MTR at 2 years was 37.5pu (SD 0.7pu) whilst at baseline MS NAWM MTR was 37.8pu (SD 0.4pu), p = 0.035. No difference was seen over time in control white matter at 2 year follow up. MS NAGM MTR at 2 years was 31.7pu (SD 0.7pu) whilst at baseline MS NAGM MTR was 32.0pu (SD 0.7pu), p = 0.015. No difference was seen over time in control grey matter. At 1 year, significant changes from baseline were also seen in MS NAGM (p < 0.001) and MS NAWM (p = 0.019), however there was also a significant increase of control white matter MTR at that time point (p = 0.028). Small yet significant difference were seen when whole white matter was compared with NAWM. Whole white matter was 0.03pu lower than NAWM. No relationship with disability was observed.

**Conclusion**

This study suggests that normal appearing grey and white matter pathology is apparent early in the course of relapsing remitting MS and that tissue specific MTR histograms are a sensitive measure of this abnormality. MTR appears capable of monitoring progressive normal appearing tissue change in a cohort who have not yet cumulated permanent disability and may potentially act as an effective surrogate marker in therapeutic trials.

**References:**


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