Magnetization transfer ratio measurement in multiple sclerosis normal-appearing brain tissue: limited differences with controls but relationships with clinical and MR measures of disease

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We investigated the magnetization transfer ratio (MTR) of normal-appearing white (NAWM) and grey matter (NAGM) in a relatively large group of multiple sclerosis (MS) patients, and the relations of MTR changes with clinical disability. MTR was measured in 66 MS patients (12 PP, 35 RR, 19 SP) and 23 healthy controls, using a whole-brain 3D-FLASH technique corrected post-hoc for B1-induced variation. Histogram parameters of conservatively selected NAWM and cortical NAGM were analysed using Bonferroni-corrected ANOVA with age as covariate. Additionally, manually outlined regions of interest were analysed using a multilevel method. Lesions had low MTR (mean 22.7 ± 6.9%), but NAWM exhibited limited changes: MTR histogram peak position was 32.8 ± 1.0% in controls and 32.4 ± 0.9% in MS patients, with a significant decrease compared to controls only in SPMS patients (31.9 ± 1.1%, p = 0.045). Cortical NAGM histograms did not differ significantly between patients and controls. In SPMS, regional mean MTR was significantly decreased in corpus callosum and hippocampus. MTR histogram parameters of NAGM and NAWM were correlated with EDSS and MSFC scores, with lesion volume and with normalized brain volume. We conclude that disease-induced MTR changes were small in MS NAWM and NAGM, but did correlate with clinical decline, lesion volume and overall cerebral atrophy. Multiple Sclerosis 2007; 13: 708–716.

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Key words: B1 correction; magnetization transfer; multiple sclerosis; normal-appearing brain tissue

Introduction

Multiple sclerosis (MS) is an inflammatory, demyelinating disease of the central nervous system characterized by lesions in the white matter. Disease effects have also been demonstrated in brain tissue not affected on conventional MR images, ie, the so-called normal-appearing brain tissue (NABT). Post-mortem studies have demonstrated astrogliosis, microglial activation, demyelination and axonal damage in normal-appearing white matter (NAWM) [1–5], and demyelinating, non-inflammatory lesions in normal-appearing grey matter (NAGM) [6,7]. In vivo, abnormalities have been demonstrated with quantitative MR techniques such as diffusion tensor imaging, T1 relaxation time mapping and MR spectroscopy (eg, [8–10]).

The magnetization transfer ratio (MTR) is a composite measure that reflects both the direct saturation effect of an off-resonance RF pulse on the free water spins, and the effect of chemical and magnetization transfer between free and bound...
spins. It is attractive because, in contrast to more fundamental parameters (eg, [11]), MTR can be readily measured on clinical MR systems within a limited time. Moreover, in combined MR–histopathology studies, decreased MTR has been related to demyelination and axonal loss in MS [4,12]. Therefore, it has been widely applied in MS research, although the influences of scanner hardware and inhomogeneity of the exciting (B1) radio-frequency (RF) field remain limitations of the method (eg, [13]). In vivo, MTR has shown evidence of local damage in NAWM prior to lesion formation (eg, [14]), and a relation between MTR decrease in NAWM and clinical decline [15].

Small but significant decreases in MTR of NAWM were found in MS patients compared to healthy controls both in regional [16,17] and global analyses [18–21]. Results on MTR of NAGM are conflicting: some studies found decreased MTR in MS and some did not [19–26]. An important note here is that, as a result of the inability of conventional MR imaging techniques to detect GM lesions, NAGM may very well contain MS lesions and is therefore likely to be heterogeneous both within and between patients.

The current study aimed to prospectively measure MTR in NAWM and NAGM in a relatively large group of MS patients. To avoid effects of slice profile imperfections and variation of the B1 RF field strength, we used a 3D gradient echo (3D-FLASH) technique with a post-hoc B1 correction of MTR values [27]. Global and regional MTR properties were compared between MS disease types and with controls, and relations with clinical disability scores and T2-lesions were explored.

Subjects and methods

Subjects

Sixty-six patients with clinically definite MS [28], divided into three groups according to clinical type (12 primary progressive [PP], 35 relapsing–remitting [RR], 19 secondary progressive [SP]) were recruited from the outpatient clinic of the MS Center specifically for this study. Twenty-three healthy volunteers without neurological disease were also recruited specifically for this study (Table 1). Patients were included in the study based on their willingness to undergo extensive MRI. Further inclusion criteria were age between 18 and 70 years, and for the MS patients no other neurological disease and no recent disease activity (relapse within previous four weeks). Of the patients with RRMS, 15 (43%) were on treatment with interferon-beta. No patients used other disease-modifying treatments. The study was approved by the institutional ethics review board. All subjects gave informed consent. The MS patients underwent a neurological examination in which expanded disability status scale (EDSS) [29] and Multiple Sclerosis functional composite (MSFC) [30] scores were determined.

MR protocol

All MR investigations were carried out with a Siemens Magnetom Vision scanner operating at 1.5 T (Siemens, Erlangen, Germany) using the standard circularly polarized transmit–receive head-coil. The MR protocol included proton density (Pd)/T2-weighted fast spin echo images (TR/TE/TE2 2625/16/98 ms, number of excitations [NEX] = 2), scanned in two interleaved sets of 16 4-mm slices each, with 1 × 1 mm² in-plane resolution. Two sets of 3D-FLASH images (TR/TE 27/4 ms, flip angle 20°, NEX = 2, 1 × 1 × 4 mm³ voxels) covering the same volume were acquired, one with an MT-prepulse (Gaussian pulse shape, 7.68 ms duration, frequency offset 1500 Hz, equivalent flip angle 500°), and one without. For the B1 calculations, five additional sets of sagittal 3D-FLASH images (“B1 3D-FLASH images”) (TR/TE 25/5 ms, NEX = 1, 2 × 2 × 4 mm³ voxels) were acquired with a non-selective pulse and nominal flip angles varying in steps of 20° between 140° and 220°.

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Subject group characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Controls</td>
</tr>
<tr>
<td>Number of subjects (M/F)</td>
<td>23 (12/11)</td>
</tr>
<tr>
<td>Age (years)</td>
<td>30.6 ± 7.4</td>
</tr>
<tr>
<td>Disease duration (years)</td>
<td>13.0 ± 9.6</td>
</tr>
<tr>
<td>Median supratentorial lesion load (range) [mL]</td>
<td>5.6 (0.2–27.8)</td>
</tr>
<tr>
<td>Normalized brain volume (L)</td>
<td>1.52 ± 0.04</td>
</tr>
<tr>
<td>Median EDSS score (range)</td>
<td>4.5 (3.0–6.5)</td>
</tr>
<tr>
<td>MSFC score</td>
<td>0.29 ± 0.19</td>
</tr>
</tbody>
</table>

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Lesions, brain volume and tissue type identification

All analyses were restricted to supratentorial brain tissue, because of the difficulties in reliably differentiating white and grey matter in the cerebellum. All image manipulation and analysis was performed using tools from FMRIB’s Software Library (FSL, www.fmrib.ox.ac.uk/fsl). For the MS patients, MR visible lesions (focal lesions and diffuse abnormalities) in the supratentorial brain were manually marked on the Pd-/T2-weighted images using in-house developed software with a semi-automated local-thresholding technique. Lesion loads were calculated from these markings. Normalized brain volume (NBV), ie, the brain volume scaled to standard head size (using the MNI-152 template), was calculated for each subject from the 3D-FLASH images without MT prepulse, using an automated procedure (SIENAX, part of FSL) with manual editing. Supratentorial white and grey matter masks were generated from the same images by the automated brain segmentation algorithm FAST (part of FSL) with a standard brain template, followed by in-plane erosion. For the MS patients, the lesion markings were co-registered with linear image registration and subtracted from the WM and GM masks to obtain conservative NAWM and NAGM masks.

MTR calculations

Using a trilinear interpolation, the 3D-FLASH images acquired with MT prepulse were registered to those acquired without, and MTR maps were generated by calculating for each pixel MTR = (M0 - M0')/M0, where M0 indicates the signal intensity without pulse, and M0' that with pulse. The B1 3D-FLASH images were spatially smoothed to increase the signal-to-noise ratio (SNR), and then a pixel-by-pixel calculation of the effective B1 RF field strength was performed by determining the nominal flip angle corresponding to a true flip angle of 180° from the signal zero crossing [31]. Using the method described by Ropele et al. [27], an individual correction for B1-induced variation was applied for each subject by deriving the relation between MTR and B1 for NAWM, and then generalizing the result to apply the correction to all tissues. Although from simulations [27], a non-linear relationship between MTR and B1 may be expected for the sequence type used in this study, our data did not show any non-linearities. Therefore, we used a linear regression to describe the relation between MTR and B1. The B1-corrected MTR maps were combined with the NAWM and NAGM masks to yield tissue-specific B1-corrected MTR maps, from which NAWM and NAGM MTR histograms with a bin size of 0.1% MTR were generated for each subject. Histograms were smoothed using a running average method, and normalized to the total pixel count to eliminate the effect of differences in NAWM or NAGM volumes between subjects. Histograms were then characterized by three parameters: peak position, peak width (full width at half maximum, FWHM) and peak height. For comparison, MTR histograms were also generated for the MR visible lesions, by eroding the co-registered lesion masks and combining them with the MTR maps. For visual presentation of the gross group behaviour, mean histograms were calculated for each subject group by averaging the unsmoothed, normalized (to the total pixel count) histograms, and smoothing the resulting average histogram.

Regions of interest (ROIs) were drawn in normal-appearing tissue on the Pd-/T2-weighted images in genu and splenium of corpus callosum, and further bilaterally in 4 WM regions, 6 cortical GM regions and 3 deep grey matter regions. After registering the ROIs to the MTR maps, the NAWM and NAGM masks were used to exclude any pixels of an inappropriate type that may have been included due to this registration step. Only ROIs consisting of at least 50 pixels (0.2 mL) after these steps were included in the analysis. As a consequence, the number of subjects with data for a region varied between 79 and 85 out of the total of 88 subjects, with a similar distribution across subject groups for all regions.

Statistical analysis

Using SPSS for Windows 12.0, histogram parameters (peak position, width and height) were compared between the combined MS group and controls using univariate analyses of variance (ANOVA’s), and also between the four subject groups using additional ANOVA’s. Subject age was included as a covariate, and the interaction between age and group if this was significant. A Bonferroni correction for multiple comparisons was applied. The regional MTR data were analysed with MlwiN [32], using a separate multilevel model for each of the three classes NAWM, cortical NAGM and deep NAGM, with subjects as level-2 units and measurement occasion as level-1 units. Each model consisted of the explanatory variables subject group, region, age, gender, the interaction between subject group and region, and the other significant first order interactions, which were identified by a model search procedure. Weights were calculated for each measurement from the size of the region and the variance of MTR as weight = size/(MTR variance), and then standardized. Two
sets of Wald $\chi^2$ tests were carried out for each model: 1) For each region, an overall test assessing differences between the four subject groups, followed, if significant, by all pairwise comparisons between groups with Bonferroni correction and a comparison between controls and MS patients. 2) The same tests as in 1) but for each class as a whole, ie, averaged over regions. In case of significant interactions (other than that between subject group and region), all the above effects were averaged with respect to the appropriate variables (ie, evaluated at the mean value of age and using effect coding for gender).

Pearson’s correlation coefficient, indicated as $r$, was used to investigate correlations of MTR histogram peak heights and peak positions with NBV and MSFC scores. Correlations with lesion loads, EDSS scores and the components of the MSFC score (ie, 9-hole peg test for dominant and non-dominant hand, paced auditory serial addition test [PASAT], 20-foot timed walk test) were investigated using Spearman’s rank correlation coefficient, indicated as $\rho$. Results are displayed as mean $\pm$ standard deviation unless indicated otherwise. Appropriately Bonferroni-corrected $P$-values less than 0.05 were considered statistically significant.

## Results

Table 1 shows the characteristics of the four subject groups. The clinical and conventional MRI data are as expected, with highest lesion loads, and lowest NBV values in the SPMS patients.

## Global analysis

Figure 1 shows the group mean MTR histograms for NAWM and NAGM. For comparison, the lesion mean MTR histogram is shown together with the NAWM histograms. Table 2 lists the group mean

![Figure 1](image-url)

**Figure 1** Group mean MTR histograms of NAWM (a) and cortical NAGM (b). Although MTR changes in lesions are substantial, as evident from the mean lesion histogram shown in (a), changes in NAWM are limited compared to control WM. MTR histograms of cortical NAGM did not reveal differences with controls.

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values of the NAWM and NAGM histogram parameters derived from individual histograms. The interaction between age and subject group was not statistically significant for any histogram parameter, so it was excluded from all ANOVA models. The results of statistical comparisons between groups are indicated in Table 2. There were no statistically significant differences between the combined MS group and controls. The NAWM MTR histogram peak position was significantly lower in SPMS than in controls (Bonferroni-corrected $P = 0.045$). The NAGM MTR histogram peak position was significantly lower in SPMS than in RRMS ($P = 0.012$). Both Figure 1 and Table 2 suggest that NAGM MTR histogram peaks of RRMS are unexpectedly shifted to slightly higher MTR values compared to controls, but this trend is not statistically significant.

### Table 3  Regional mean MTR values

<table>
<thead>
<tr>
<th>Region</th>
<th>Controls</th>
<th>PPMS</th>
<th>RRMS</th>
<th>SPMS</th>
<th>Combined MS group</th>
</tr>
</thead>
<tbody>
<tr>
<td>White matter</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Internal capsule</td>
<td>$33.0 \pm 1.3$</td>
<td>$33.4 \pm 1.1$</td>
<td>$33.1 \pm 1.3$</td>
<td>$32.5 \pm 1.1$</td>
<td>$33.0 \pm 1.2$</td>
</tr>
<tr>
<td>Centrum semi-ovale</td>
<td>$32.5 \pm 1.2$</td>
<td>$32.2 \pm 1.1$</td>
<td>$32.0 \pm 1.4$</td>
<td>$31.7 \pm 1.2$</td>
<td>$32.0 \pm 1.3$</td>
</tr>
<tr>
<td>Genu of corpus callosum</td>
<td>$35.1 \pm 1.1$</td>
<td>$34.4 \pm 0.6$</td>
<td>$34.5 \pm 1.7$</td>
<td>$33.4 \pm 1.7^*$</td>
<td>$34.2 \pm 1.6^{**}$</td>
</tr>
<tr>
<td>Splenium of corpus callosum</td>
<td>$34.0 \pm 1.2$</td>
<td>$34.0 \pm 1.3$</td>
<td>$35.3 \pm 1.5$</td>
<td>$32.8 \pm 1.2^*$</td>
<td>$33.3 \pm 1.4$</td>
</tr>
<tr>
<td>Frontal NAWM</td>
<td>$33.7 \pm 1.0$</td>
<td>$33.4 \pm 0.9$</td>
<td>$33.5 \pm 1.2$</td>
<td>$33.1 \pm 1.4$</td>
<td>$33.3 \pm 1.2$</td>
</tr>
<tr>
<td>Parieto-occipital NAWM</td>
<td>$32.6 \pm 0.8$</td>
<td>$32.6 \pm 0.7$</td>
<td>$32.4 \pm 1.1$</td>
<td>$32.3 \pm 1.1$</td>
<td>$32.4 \pm 1.0$</td>
</tr>
<tr>
<td>Cortical gray matter</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cingulate cortex</td>
<td>$23.5 \pm 1.2$</td>
<td>$23.8 \pm 1.2$</td>
<td>$23.9 \pm 1.5$</td>
<td>$22.9 \pm 1.2$</td>
<td>$23.6 \pm 1.4$</td>
</tr>
<tr>
<td>Frontal cortex</td>
<td>$24.5 \pm 1.0$</td>
<td>$24.5 \pm 1.7$</td>
<td>$24.5 \pm 1.5$</td>
<td>$24.4 \pm 1.1$</td>
<td>$24.5 \pm 1.4$</td>
</tr>
<tr>
<td>Hippocampus</td>
<td>$22.7 \pm 1.3$</td>
<td>$21.5 \pm 2.0$</td>
<td>$22.9 \pm 1.8$</td>
<td>$21.1 \pm 2.0^*$</td>
<td>$22.2 \pm 2.0$</td>
</tr>
<tr>
<td>Insular cortex</td>
<td>$22.5 \pm 0.9$</td>
<td>$23.0 \pm 2.0$</td>
<td>$23.0 \pm 1.8$</td>
<td>$22.6 \pm 1.4$</td>
<td>$22.9 \pm 1.7$</td>
</tr>
<tr>
<td>Parieto-occipital cortex</td>
<td>$23.4 \pm 1.0$</td>
<td>$23.6 \pm 0.8$</td>
<td>$23.6 \pm 1.2$</td>
<td>$22.9 \pm 1.2$</td>
<td>$23.4 \pm 1.2$</td>
</tr>
<tr>
<td>Striate cortex</td>
<td>$22.5 \pm 1.6$</td>
<td>$21.9 \pm 0.8$</td>
<td>$22.8 \pm 1.5$</td>
<td>$21.3 \pm 1.3^*$</td>
<td>$22.2 \pm 1.5$</td>
</tr>
<tr>
<td>Deep gray matter</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caudate nucleus</td>
<td>$25.0 \pm 1.3$</td>
<td>$24.9 \pm 1.1$</td>
<td>$25.4 \pm 1.3$</td>
<td>$24.8 \pm 2.4$</td>
<td>$25.1 \pm 1.7$</td>
</tr>
<tr>
<td>Putamen</td>
<td>$25.9 \pm 1.3$</td>
<td>$25.8 \pm 1.4$</td>
<td>$26.3 \pm 1.3$</td>
<td>$25.6 \pm 1.5$</td>
<td>$26.0 \pm 1.4$</td>
</tr>
<tr>
<td>Thalamus</td>
<td>$28.4 \pm 1.4$</td>
<td>$28.7 \pm 1.2$</td>
<td>$29.3 \pm 1.1$</td>
<td>$28.0 \pm 1.2^{**}$</td>
<td>$28.8 \pm 1.2$</td>
</tr>
</tbody>
</table>

*Values are shown as means and standard deviations. Bonferroni-corrected $P$-values from multilevel analyses are indicated: $^*P < 0.05$ compared to controls, $^{**}P < 0.01$ compared to controls, $^{*}P < 0.05$ compared to RRMS, $^{**}P = 0.001$ compared to RRMS.

### Regional analysis

In all NAWM regions the data show a tendency for lower MTR values in MS patients than in controls (Table 3). This was not found for grey matter, where there was substantial variation within groups. Significant interactions were that between gender and age for deep NAGM, and that between gender and region for NAWM. These interactions were included in the appropriate models. Comparing the regional MTR values of MS patients to those of controls, the only significant MTR decrease ($P = 0.009$) was found in the genu of the corpus callosum, while trends ($P$ values between 0.051 and 0.053) were found for the splenium of the corpus callosum, centrum semi-ovale and hippocampus. In the mutual comparisons of the four subject groups, the decrease in MTR
in SPMS compared to controls was significant in the combined class of NAWM ROIs ($P = 0.049$), and also in two individual NAWM ROIs, namely in the genu ($P = 0.013$) and splenium ($P = 0.015$) of the corpus callosum, while a trend was observed for the centrum semiovale ($P = 0.075$). In the hippocampus, MTR was significantly lower in SPMS than in controls ($P = 0.014$). MTR was significantly lower in SPMS than in RRMS in thalamus ($P = 0.001$), hippocampus ($P = 0.012$) and striate cortex ($P = 0.014$), while there was a corresponding trend in the combined class of cortical NAGM ROIs ($P = 0.063$).

Correlations

Within the group of MS patients, peak positions were correlated with MSFC scores (NAWM Pearson’s $r = 0.347$, $P = 0.007$; NAGM $r = 0.460$, $P < 0.001$) and with EDSS scores (NAWM Spearman’s $\rho = -0.268$, $P = 0.032$; NAGM $\rho = -0.302$, $P = 0.015$). Figure 2 displays a scatter plot of MSFC score versus NAGM MTR histogram peak position. There were no correlations with MSFC or EDSS scores for peak heights.

We also investigated the correlations between the separate components of the MSFC score and the MTR histogram parameters. PASAT scores did not correlate with any histogram parameter. MTR peak height and peak position of NAWM were significantly negatively correlated with nine-hole peg test (9HPT) scores for both dominant and non-dominant hand, with the strongest correlation between 9HPT of the dominant hand and NAWM MTR peak height ($\rho = -0.425$, $P = 0.001$). NAGM MTR parameters showed similar behaviour, but NAGM MTR peak height did not correlate significantly with 9HPT of the non-dominant hand ($\rho = -0.210$, $P = 0.102$). The strongest correlations for NAGM were observed for NAGM peak height versus 9HPT of the dominant hand ($\rho = -0.417$, $P = 0.001$), and for NAGM peak position versus 9HPT of the non-dominant hand ($\rho = -0.423$, $P = 0.001$). The 20-foot timed walk test score correlated only with NAGM peak position ($\rho = -0.306$, $P = 0.015$).

MTR histogram parameters were correlated with brain volume for MS patients, but not for controls. For the MS patients, all peak heights and peak positions of NAWM and NAGM were significantly correlated with NBV (all $r > 0.40$, $P < 0.001$ except NAGM peak height: $r = 0.271$, $P = 0.031$).

Lesion load of the MS patients was correlated significantly with all MTR peak heights and positions, most strongly with NAWM peak height ($\rho = -0.487$, $P < 0.001$) and NAGM peak position ($\rho = -0.466$, $P < 0.001$).

Discussion

We observed small global and regional MTR decreases in MS NAWM compared to control WM, comparable in size to the MTR changes found in other studies [16–21,26]. Although differences between groups were small, the NAWM and NAGM MTR histogram parameters correlated with WM lesion volume, with NBV and with clinical disability in the group of MS patients.

Some previous studies of MTR in MS have reported slightly larger NAWM MTR changes than those observed in the present work, and all previous studies of whole-brain NAWM have reported peak position shifts reaching statistical significance, whereas this was not the case in our study [18–21]. This discrepancy of our study with other studies did not arise as a result of the post-hoc correction for $B_1$ imperfections, because, in accordance with numerical simulations [27], the variation of MTR within the $B_1$ error range in this study was limited and the correction therefore had only a small effect. The discrepancy is also not caused by the co-registration of images with and without MT-prepulse, because that was also used in three of the four previous studies, with comparable voxel sizes. Possibly, our technique may be less sensitive to subtle pathological changes than others, although it seems unlikely in light of the fact that the MTR decrease in lesions in this study is large compared to that reported by others [20,33,34]. The lower mean age of the group of healthy controls could be responsible for the absence of the expected difference between MS patients and controls. Three studies have investigated the age-dependence of MTR: two found no relation with age in the age range of the subjects studied here [35,36], and another study found significant but
small differences between subjects under and over 35 years of age [37], but these indicate that younger subjects should be expected to have higher MTR values, which would artificially enhance the disease effect in our study. For these reasons, the difference in age is not expected to influence our results. Nevertheless, to deal with this issue, age was included as a covariate in all ANOVA analyses of histogram parameters.

It is a common finding of our study and other studies [16,17,26] that MTR decreases in MS NAWM are small compared to those in MS lesions. This suggests that demyelination and axonal loss, features that MTR is thought to represent [4,12], occur only to a limited extent in MS NAWM, or involve only a limited fraction of the NAWM. Histopathological investigations have demonstrated axonal damage and loss in cerebral NAWM [2,3,5], but also only to a small extent. For example, Trapp et al. report that the number of transected axons per mm$^3$ (mean ± standard error) is 17 ± 2.8 in non-lesion MS white matter versus 0.7 ± 0.7 in control white matter, while in active MS lesions the number is 11.236 ± 2775 [5]. The extent of the axonal damage in NAWM is thus so small that it may well only give rise to small MTR decreases. The pronounced global changes reported for $T_1$ apparent diffusion coefficient (ADC) and diffusion fractional anisotropy (FA) [8,9,38,39] may therefore mainly result from oedema, glial proliferation or other processes that do not inflict actual tissue damage. This would be in line with observations of MS NAWM from localized long repetition time, short echo time proton MR spectroscopy that suggest an increase of glial cells (increase of myoinositol and creatine) but no neuro-axonal damage (normal N-acetyl-aspartate) [10,40].

In deep grey matter no differences were found between patients with MS and healthy controls in regional mean MTR, in line with Filippi et al. [23], although they did observe trends. Audoin et al. did observe significantly lower MTR in thalamus in MS compared to controls using a voxel-based method [22]. In our study, we did observe MTR differences in thalamus, but not with controls: the only significant deep grey matter MTR difference was between SPMS and RRMS in thalamus.

In our study, MTR of cortical NAGM did not differ significantly between MS patients and controls. We observed no significant regional MTR decrease in MS compared to controls, and neither did previous studies [25,26]. We also did not find significant changes in MTR histogram parameters of cortical NAGM, whereas two previous studies found a significantly lower NAGM peak position in MS compared to controls [19,22], but three other studies did not [20,21,24]. Previous histopathological studies have demonstrated the occurrence of focal demyelinating cortical grey matter lesions in MS [6,7], and these lesions may account for the observations of neuro-axonal damage in cortical NAGM in patients with SPMS [41,42]. In light of these histopathological and MR spectroscopic observations, the MTR findings raise the question whether MTR is a suitable measure for detecting MS disease processes or damage in cortical NAGM. One could argue that when using MTR, decreases in myelin content may be much harder to detect in cortical grey matter than in white matter, because cortical grey matter has relatively low myelin content leading to MTR values that are already low in healthy tissue, and because cortical grey matter has a lower SNR than white matter on the T1-weighted images generally used in the measurement of MTR.

However, there is some evidence of the contrary. First, MTR of cortical NAGM was lower in SPMS than in RRMS in our study, globally, and also regionally in hippocampus and striate cortex, which is in line with expectations of greater cortical damage in SPMS. Second, in the group of patients with MS, cortical NAGM MTR histogram parameters were correlated with clinical disability scores, with NBVs, and with lesion loads. Although there were no significant differences between MS patient groups and the control group, these correlations with other measures of disease suggest that MTR does detect effects of MS disease processes in cortical NAGM. Moreover, several recent studies using MTR have found that cortical MTR is related to disability, and even predictive of future accumulation of disease [43–45]. This supports the view that MTR can measure disease effects in MS cortical grey matter.

Correlations with MSFC and EDSS scores were relatively weak. Slightly stronger negative correlations with histogram peak positions and heights were observed for 9HPT scores. Interpreting these as overall measures of cortical GM integrity, the correlations with 9HPT scores suggest that the degree to which the upper extremity function is diminished is related to the degree of overall cortical degradation. This may be either because damage in most parts of the cortex will have some effect on 9HPT performance, or because the degree of damage in regions relevant to the task, eg, in the sensorimotor cortex or visual cortex, is generally related to the degree of damage in other cortical areas. In support of the latter argument, previous studies have suggested that normal-appearing grey matter damage may be a widespread process affecting regions throughout the brain [39,46–48].

The regional data revealed significantly decreased MTR in MS compared to healthy controls only in corpus callosum and hippocampus, implying that throughout the NABT, changes in MTR are

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limited. As MTR decrease is supposed to reflect mainly demyelination and axonal loss, this suggests these processes may occur only to a limited degree in nearly all cerebral structures. The correlations of overall NAGM MTR parameters with clinical scales demonstrate that, although the changes may be not or only just detectable with MTR, they are clinically relevant. As argued above, MTR measurements may not be the most sensitive method of investigating cortical changes in MS. Other techniques, such as regional volume measurements [46], voxel-based morphometry [49] and cortical thickness measurements [47], combined with developments aimed at improving MR imaging sensitivity to focal lesions [50], may be more suitable for detecting the cortical changes in MS.

In conclusion, the differences between patients with MS and healthy controls in MTR of both NAWM and NAGM were small in our study. However, within the MS patient group, the MTR histogram parameters of both NAWM and cortical NAGM did exhibit a relationship with clinical disability scores, with brain atrophy and with the volume of MR-visible lesions. This implies that although MTR changes may be small, they do reflect clinically relevant changes.

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References


