MULTIPLE SCLEROSIS JOURNAL | MSJ

Original Research Paper

Metabolites predict lesion formation and severity in relapsing-remitting multiple sclerosis

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Multiple Sclerosis Journal

2018, Vol. 24(4) 491–500

DOI: 10.1177/1352458517702534

Abstract

Background: Multiple sclerosis is characterized by white matter lesions, which are visualized with conventional T2-weighted magnetic resonance imaging (MRI). Little is known about local metabolic processes preceding the appearance and during the pathological development of new lesions.

Objective: To identify metabolite changes preceding white matter (WM) lesions and pathological severity of lesions over time.

Methods: A total of 59 relapsing-remitting multiple sclerosis (MS) patients were scanned four times, with 6-month intervals. Imaging included short-TE magnetic resonance spectroscopic imaging (MRSI) and diffusion tensor imaging (DTI).

Results: A total of 16 new lesions appeared within the MRSI slab in 12 patients. Glutamate increased (+1.0 mM (+19%), $p = 0.039$) 12 and 6 months before new lesions appeared. In these areas, the increase in creatine and choline 6 months before until lesion appearance was negatively correlated with radial diffusivity ($\rho = -0.73$, $p = 0.002$ and $\rho = -0.72$, $p = 0.002$). Increase in creatine also correlated with the increase of axial diffusivity in the same period ($\rho = -0.53$, $p = 0.034$). When splitting the lesions into "mild" and "severe" based on radial diffusivity, only mild lesions showed an increase in creatine and choline during lesion formation ($p = 0.039$ and $p = 0.008$, respectively).

Conclusion: Increased glutamate heralded the appearance of new T2-visible WM lesions. In pathologically "mild" lesions, an increase in creatine and choline was found during lesion formation.

Keywords: Multiple sclerosis, MRI, MRS, MRSI, DTI, metabolite

Date received: 19 October 2016; revised: 16 February 2017; accepted: 8 March 2017

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Introduction

Multiple sclerosis (MS) is an inflammatory, neurodegenerative disease that is characterized by focal white matter (WM) lesions, among other pathological changes.[1] Conventional T2-weighted magnetic resonance imaging (MRI) is an essential tool for diagnosis although the number of T2 WM lesions is a poor predictor of clinical disability.[2] More advanced MRI techniques are needed to be able to detect varying degrees of (subtle) tissue damage within lesions or in the normal-appearing white matter (NAWM), to better explain clinical disability. Magnetic resonance spectroscopic imaging (MRSI) and diffusion tensor imaging (DTI) are two such techniques, giving information about metabolism and tissue microstructure, respectively.[3–7]

One previous study investigated metabolite changes prior to lesion appearance in MS[8] and found an increased choline/creatine ratio in WM areas that later showed lesions. Metabolite changes during lesion formation have also been sparsely studied.[9–13] A few studies reported increases in total creatine and choline or a transient decrease in total *N*-acetylaspartate or its ratio to creatine when following acute, gadolinium-enhancing lesions over time. The relation between metabolite changes and microstructural tissue damage over time has not been investigated before.

In the present longitudinal study, we aimed to identify local metabolite changes both prior to and during WM lesion formation, using short echo-time MRSI. Changes in metabolite concentrations were related to changes in

DTI-measured tissue damage in lesions, in order to study the predictive value of early metabolite changes in terms of lesion development and tissue damage.

Materials and methods

Study design

A total of 59 relapsing-remitting MS patients were included in this study and scanned four times, with a 6-month interval. The study is exploratory and no a priori sample size was estimated. Data, part of the same group, have been published previously.[14,15] In total 29 patients were starting natalizumab, 19 patients were continuing interferon or glatiramer acetate, and 11 patients were not using disease-modifying-drugs at the baseline measurement. Inclusion criteria were a diagnosis of clinically definite relapsing-remitting MS[16] and an age between 18 and 65 years. Patients with a history of neurological or psychiatric conditions (besides relapsing-remitting MS), or a history of alcohol or drug abuse were excluded.

Standard protocol approvals, registrations, and patient consents

The research protocol was approved by the institutional ethics review board, and written informed consent was given by all subjects before participation.

MRI acquisition

MRI was performed on a 1.5T whole-body scanner (Siemens Sonata, Erlangen, Germany) with an eight-channel receiver head coil.

Metabolite spectra were measured with a two-dimensional (2D) MRSI, point resolved spectroscopy (PRESS) sequence (repetition time (TR)/echo time (TE): 3000/30 ms) on a 15-mm thick slab, aligned to the pituitary-fastigium line, with the center touching the top of the corpus callosum (Figure 1(a)). Based on head size, the field-of-view was 160 × 160 or 140 × 160 mm, and the corresponding volume of interest (VOI) was 80 × 100 or 70 × 100 mm. Within subjects, VOI was kept constant over time. The use of 16 × 16 phase-encodings resulted in a voxel size of 1.5 or 1.3 mL. Reference 2D MRSI datasets were acquired without water suppression using both head and body coils as receiver. The phase-encoded acquired data were filtered with a hamming function to reduce the outside voxel volume contamination.

DTI was performed by a diffusion-weighted echo-planar acquisition (TR: 8.500 ms; TE: 86 ms and isotropic resolution, 2 × 2 × 2 mm) including 60 volumes with noncollinear diffusion gradients (*b* value of 700 s/mm^2) and 10 volumes without directional weighting.

Structural imaging included a three-dimensional (3D) T1-weighted magnetization prepared rapid acquisition gradient-echo (MPRAGE) (TR: 2700 ms; TE: 5 ms; inversion time (TI): 950 ms; 176 sagittal slices; 1.3 mm section thickness; 1.3 × 1.3 mm^2 in-plane resolution), and a Pronton density/T2-weighted turbo spin-echo (TR: 3130 ms; TE: 24 and 85 ms; 46 contiguous 3 mm axial slices; 1 × 1 mm^2 in-plane, aligned to the 2D MRSI slab). No T1-weighted contrast-enhanced imaging was performed.

Lesion masks

When a new lesion appeared in the MRSI slab during the study period, a lesion mask, that is, a 3D volume delineating a region of interest, was manually created on the corresponding T2-hyperintensity. New T2-hyperintense MS lesions were assessed by experienced neuroradiologists and only convincing new lesions were retained for manual segmentation. The lesion mask was then registered onto the exact same areas of the preceding timepoints, using FLIRT software.[17] The WM surrounding the lesional area was also included, by dilating all lesion masks by 18 mm. This reduced variability by including more MRSI voxels in the lesional masks. As a result, magnetic resonance spectroscopic imaging lesion masks (MRSI-LM) were created at the appearance of the new T2 lesion (0 mp), at 6 months prior to appearance (6 mp) and at 12 months prior to appearance (12 mp) (Figure 1(c)). WM lesions already visible at the first acquisition were dilated in-plane by 2 mm and included in a chronic lesion mask. Because this study focused on the metabolite changes preceding and during WM lesion formation, metabolite concentrations in chronic lesion mask were dismissed and masked out. More details are given in the online Supplementary Material.

Metabolite quantification

The spectrum of each MRSI voxel was quantified using LCModel[18] resulting in absolute concentrations of *N*-acetylaspartate and *N*-acetyl aspartylglutamate (tNAA), total creatine (tCr), choline-containing compounds (Cho), myo-inositol (Ins), and glutamate (Glu) expressed in mmol/liter (mM). Glutamine levels were also quantified and checked in order to ensure a reliable Glu quantification (details given in the online

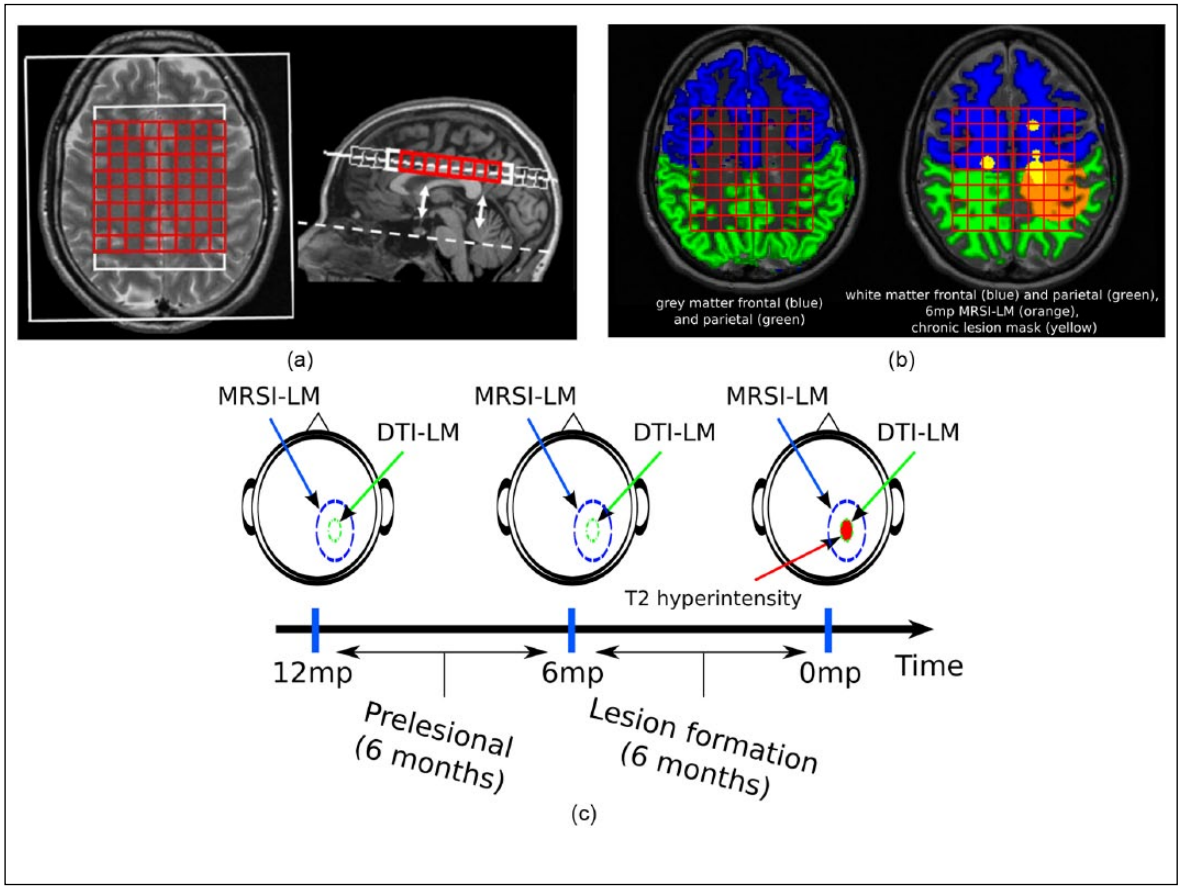

Figure 1. (a) Axial T2-weighted and sagittal T1-weighted MR images with the MRSI slab positioned in the center of the brain, aligned with the pituitary-fastigium line. On the axial image (left), the outer and inner white rectangles represent, respectively, the MRSI field-of-view and the PRESS volume-of-interest. The red grid depicts the 64 voxels retained for the analysis. (b) Based on the metabolite concentrations estimated by LCModel in every voxel of the slab, post-processing was performed to determine concentrations in the several MRSI lesion masks (MRSI-LM) taking into account the contribution of WM and GM and chronic lesions (see text for specific definitions). (c) Based on the appearance of a new lesion as T2-hyperintensity (red), a set of MRSI lesion masks (MRSI-LM) (dashed blue) and DTI lesion masks (DTI-LM) (dashed green) were defined at the appearance of the new T2 lesion (0 pm), at the timepoint 6 months prior to appearance (6 mp) and at the timepoint 12 months prior to appearance (12 mp). Two periods of time are also defined corresponding to the successive lesion formation stages: the pre-lesional period and the lesion formation period.

Supplementary Material). Quality assessment and reproducibility of the absolute metabolite quantification for these metabolites have been published previously.[14] See online Supplementary Material for full details.

Concentrations in lesion masks

The 3D T1 images were lesion-filled using LEAP software[20] and segmented using SIENAX software,[17] producing gray matter (GM) and WM masks. Overlaps with MRSI-LMs were removed resulting in GM and NAWM masks. These were then divided in frontal and parietal lobe sub-masks based on the *ICBM 152* nonlinear 2009c atlas (Figure 1(b)).[19] More details about the steps are described in the online Supplementary Material.

Using the collective of these masks, brain tissue within the MRSI slab was divided into MRSI-LMs at 0, 6, and 12 mp, chronic lesion mask, frontal gray or NAWM, and parietal gray or NAWM. Metabolite concentrations were then estimated within each area by an extrapolation analysis. This post-processing method extrapolates concentrations within all the masks depending on their partial volume contributions to each MRSI voxel. Changes in metabolite concentrations in MRSI-LM were calculated for every 6-month period between two acquisitions (Figure 1(c)): the pre-lesional period (differences between 12 and 6 mp in MRSI-LM) and the lesion formation period (differences between 6 and 0 mp in MRSI-LM). Additional details are given in the online Supplementary Material.

DTI within lesions

Diffusion tensor images were fitted to a tensor model resulting in 3D volumes of axial diffusivity (AD) and radial diffusivity (RD) in 10^{-5} mm^2/s, using DTIFIT software.[17] Diffusion tensor imaging lesion masks (DTI-LM) were delineated identically to the MRSI-LMs (Figure 1(c)) but not dilated (due to the higher spatial resolution and signal-to-noise ratio (SNR) of the DTI measurements). The mean values for AD and RD were then calculated inside DTI-LM at 12, 6, and 0 mp. The DTI metric changes in DTI-LM between the different timepoints were then calculated in the same way as for the metabolite concentrations leading to ΔAD and ΔRD during the pre-lesional period (differences between 12 and 6 mp) and the lesion formation period (differences between 6 and 0 mp). AD and RD are known to increase markedly with the formation of a new WM lesion[21] and the level of RD found in new, enhancing lesions is thought to reflect the severity of the tissue damage.[22,23] Hence, the amount of ΔRD during the lesion formation period was considered as a degree of lesion severity.

DTI changes over time and the classification of lesions as "mild" or "severe"

To explore the change in DTI metrics occurring in the lesions, the significance of the changes of ΔAD and ΔRD was assessed during the pre-lesion and lesion formation periods. To identify possible metabolite markers specific to a lesion severity subtype, two subgroups of lesions were created. The lesions above the median ΔRD were defined as "severe" and the lesions below the median as "mild."

Metabolite concentration changes during the pre-lesional period

To investigate whether and which metabolic changes occur before the appearance of new WM lesions, we measured the changes in concentrations during the pre-lesional period in the MRSI-LMs. If a metabolite showed a pre-lesional change, the same test was performed, as surrogate analysis, on the corresponding changes in concentration ratio with respect to tCr and additionally with MRSI-LM at 12 and 6 mp mirrored in the contralateral hemisphere. Finally, the concentration changes in NAWM were also analyzed to check their stability.

As a posteriori analysis, the correlation was calculated between the significant changes in metabolite concentrations in the pre-lesional period and the changes in DTI metrics during the lesion formation period and the volume of the newly formed lesions.

Metabolite concentration changes during the lesion formation period and relations with lesion severity

The significance of the metabolite concentrations changes during the lesion formation period was determined either in all the lesions combined or in the "mild" and "severe" subgroups.

In an a posteriori analysis, the correlation between metabolite concentrations exhibiting significant changes and ΔAD and ΔRD during the same time period was assessed. Correlation coefficients were also computed between concentration changes during the lesion formation period and the volume of the newly formed lesions.

Statistical analysis

Statistical longitudinal assessments of metabolite concentrations were performed using a Wilcoxon signed-rank test between the applicable timepoints. The DTI metrics change were statistically analyzed with a Student's *t*-test on paired data of the applicable timepoints. Correlations coefficients were computed using the Spearman's rank correlation. The tests were considered significant when the p value was <0.05 (values are given as mean ± standard deviation, unless indicated otherwise). Due to the exploratory nature of the study, no adjustment for multiple comparisons was made.

Results

Subjects

A total of 12 patients showed one or more formations of new lesions within the MRSI slab. These patients had a mean age of 37 years (range: 20−50 years) and mean disease duration of 4.6 years (range: 0.16−13.3 years) at first acquisition. Only data of these subjects were retained for further analysis.

Five patients received interferon beta-1b or glatiramer acetate, whereas seven patients were untreated. Mean (standard deviation) normalized brain volume, normalized WM volume, and normalized GM volume[17] were, respectively, 1.39 L (±0.07 L), 0.68 L (±0.04 L), and 0.72 L (±0.04 L). The median T2 lesion load was 4.3 mL (interquartile range: 2.1−11.9 mL). Median Expanded Disability Status Scale (EDSS) was 2.0 (range: 1−3.5). Spectral qualities of the VOI in the MRSI slab were comparable to a previous reproducibility study,[14] with a mean full-width at half-maximum (FWHM) of 5.18 Hz (±0.81 Hz) and a mean SNR of 10.8 (±1.35).

Table 1. Changes in metabolite concentrations and DTI metrics.

Average change (SD) in MRSI-LM (mM)	Pre-lesional period All lesions ($N = 9$)	Lesion formation period		
		All lesions ($N = 16$)	Mild ($N = 8$)	Severe ($N = 8$)
ΔtCr	0.00 (0.46), +0%	0.39 (0.90), +9%	0.89 (0.86), +20%, **($p = 0.039$)**	−0.10 (0.66), −2%
ΔtNAA	0.33 (0.93), +4%	0.27 (1.24), + 4%	0.78 (1.24), +10%	−0.25 (1.07), −3%
ΔCho	−0.04 (0.19), −2%	0.16 (0.32), + 11%	0.35 (0.24), +24%, **($p = 0.008$)**	−0.03 (0.27), −2%
ΔIns	0.24 (0.6), +6%	0.45 (1.21), +11%	0.89 (1.34), +21%	0.01 (0.96), 0%
ΔGlu	1.00 (1.30), +20%, **($p = 0.039$)**	0.16 (1.97), +3%	1.24 (1.99), +24%	−0.79 (1.46), −16%
Average change (SD) in DTI-LM ($10^{-5}mm^2/s$)	Pre-lesional period All lesions ($N = 9$)	Lesion formation period All lesions ($N = 16$)		
ΔAD	0.22 (1.37), +0%	6.16 (5.46), +6%, **($p = .001$)**		
ΔRD	−0.42 (1.96), −1%	10.19 (4.08), +16%, **($p < .001$)**		

SD: standard deviation; DTI: diffusion tensor imaging; MRSI-LM: magnetic resonance spectroscopic imaging lesion masks; tCr: total creatine; tNAA: *N*-acetylaspartate and *N*-acetyl aspartylglutamate; Cho: choline-containing compounds; Glu: glutamate; DTI-LM: diffusion tensor imaging lesion masks; AD: axial diffusivity; RD: radial diffusivity.
Top table: Mean (SD) change of the metabolite concentrations in the two 6-month periods with significant results displayed with the respective *p* values. The columns 2 and 3 show group results including all the lesions. In columns 4 and 5, the results of two subgroups of lesions, "mild" and "severe," that were divided around the median RD increase during the lesion formation period are displayed. Bottom table: Mean (SD) change of the DTI metrics measured in the lesions during the two 6-month periods with significant results shown with the respective *p* values.

In total, 16 newly forming WM lesions were monitored over time. Data in the pre-lesional period were only available for nine lesions (seven lesions appeared at the second acquisitions) and lesion formation period data were available for the entire 16 lesions. AD and RD did not change during the pre-lesional period but increased during the lesion formation period (online Supplementary Material, Table 1).

Pre-lesional period: glutamate increase

An increase of +19% on average ($p = 0.039$) was measured for Glu during the pre-lesional period ($\Delta Glu = 1.0 \pm 1.3$ mM) (Figure 2(a), Table 1). Other metabolites remained stable within the pre-lesional period and Glu in the contralateral (control) region also showed no changes. Analysis of the change of concentration ratio Δ(Glu/tCr) confirmed the pre-lesional increase, on average of +23% ($p = 0.027$, $\Delta(Glu/tCr) = 0.26 \pm 0.31$). The metabolite concentrations in NAWM did not show any significant changes in both the pre-lesion and the lesion formation period (Table e-1).

In the post hoc analysis, pre-lesional ΔGlu was associated with ΔAD of the lesion formation period ($\rho = 0.86$; $p < 0.003$) (Figure 2(b)); pre-lesional ΔGlu was also associated with the volume of the new lesion ($\rho = 0.68$; $p = 0.050$) (Figure 2(c)) but not with ΔRD of the lesion formation period (Table 2).

Lesion formation period: increase of tCr and Cho in developing "mild" lesions

During the lesion formation period, only the subgroup of "mild" lesions showed increases in metabolite concentrations: tCr was found to increase by 23% ($p = 0.039$, $\Delta tCr = 0.89 \pm 0.86$ mM) and Cho by 27% ($p = 0.008$, $\Delta Cho = 0.35 \pm 0.24$ mM) (Table 1).

In the a posteriori analysis, the values of ΔtCr and ΔCho in all lesions during the lesion formation period were strongly associated with ΔAD and ΔRD in the same period. ΔtCr was anti-correlated to ΔRD ($\rho = -0.73$, $p = 0.002$) and ΔAD ($\rho = -0.53$, $p = 0.034$). ΔCho was anti-correlated with ΔRD ($\rho = -0.72$, $p = 0.002$) (Figure 3(c) and (d)). ΔtCr and ΔCho were not correlated to the lesion volume (Table 2).

Discussion

This study investigated related metabolite and diffusion changes prior to and during new WM lesion formation in MS. Glutamate increases were found to

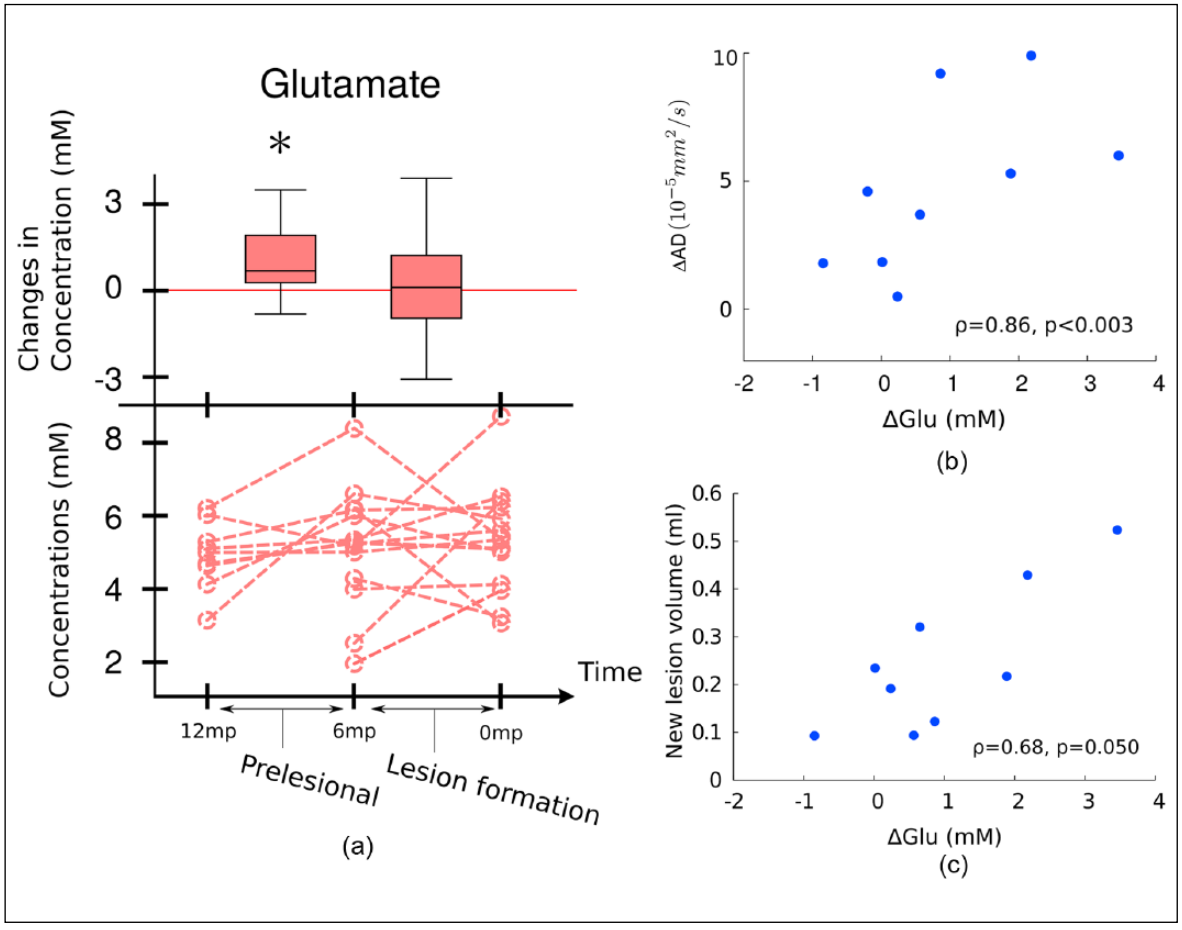

Figure 2. (a) Absolute Glu concentrations in MRSI-LM over time showed an increase in the pre-lesional period. Top: The changes in concentration (differences between two consecutive absolute concentrations) are shown (first to third quartile box, 95% span whiskers) for the pre-lesion and the lesion formation periods. The concentration changes measured during the pre-lesional period (*) were significantly greater than zero ($p = 0.039$). Bottom: The absolute concentrations measured at the three time points are displayed with each lesion represented by a line. (b) Glu increases during the pre-lesional period show a significant correlation with the change of AD during lesion formation and (c) Glu rises during the pre-lesional period were significantly correlated with the volume of the new lesion.

Table 2. Correlations between metabolite concentrations and DTI metrics.

Spearman's ρ		Lesion formation period		New lesion volume
		ΔAD	ΔRD	
Pre-lesional period	ΔGlu	0.86 (***p* = 0.003**)	0.32	0.68 (***p* = 0.050**)
Lesion formation period	ΔtCr	−0.53 (***p* = 0.034**)	−0.73 (***p* = 0.002**)	−0.10
	ΔCho	−0.35	−0.72 (***p* = 0.002**)	0.10

DTI: diffusion tensor imaging; tCr: total creatine; Cho: choline-containing compounds; Glu: glutamate; AD: axial diffusivity; RD: radial diffusivity.
Spearman's rank correlation coefficients between metabolite concentration changes and the DTI metrics changes. The last column shows the correlation coefficients between the volume of the new lesions and the metabolite concentration changes. The *p* value is shown when the correlation is significant.

precede the development of new WM lesions on T2-weighted MRI scans. Furthermore, tCr and Cho increases were associated with lower increases of RD and AD in the developing lesions. And finally, pre-lesional Glu increase was indicative of higher subsequent lesion volumes and AD increases.

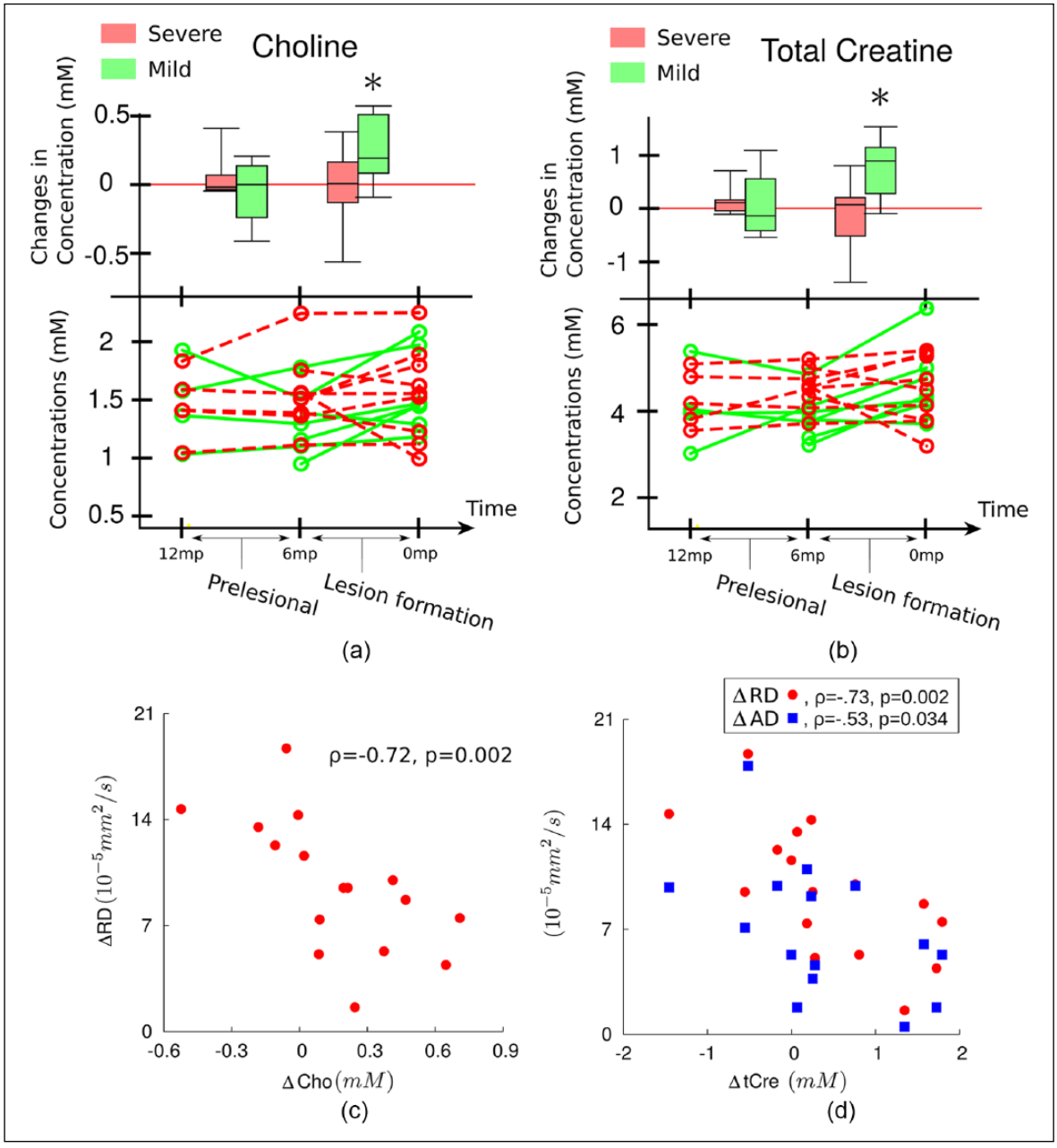

Figure 3. (a) and (b) Changes in concentration of Cho and tCr during lesion formation for the severe (red) and the mild (green) lesions are displayed in box-and-whisker (95% span). Successive measurements of (c) Cho and (d) tCr in MRSI-LM illustrate the variability in the absolute concentrations. During the lesion formation period only mild lesions displayed significant increases of Cho and tCr (*) (respectively, $p = 0.01$ and $p = 0.02$). Considering all the lesions, the increases of RD during lesion formation showed anti-correlation with the change of (c) Cho and of (d) tCr, all during lesion formation. Increases of AD during lesion formation were also correlated to (d) tCr but to a lower extent.

Pre-lesional period

Although a higher level of glutamate in NAWM and in acute WM lesions has been previously reported,[7,24] the observation of Glu rising prior to WM lesion appearance is new and important. The rise in Glu could be attributed to activated macrophages and/or microglia, which are known to cluster in the pre-lesional stages[25,26] and to produce Glu.[27] Other explanations might be subtle pathology of axons or abnormalities of Glu receptor/transporter biology leading to abnormal parenchymal Glu concentrations.[28]

The Glu increase during the pre-lesional period was strongly correlated with the AD increase, which is

generally deemed to relate to axonal injury.[4,5,29–31] The early pathological changes discussed above may therefore be indicative of the extent of eventual axonal damage within newly developing WM lesions in MS. Moreover, a stronger increase of Glu in pre-lesional WM was also associated with higher subsequent lesion volumes. Although there is an obvious effect of the lesion volume on the metabolite estimated within MRSI-LMs (considering the dilation of the lesion mask, the relative amount of actual T2-hyperintense tissue in MRSI-LM would be greater for larger lesions than for smaller lesions), the correlation in Figure 2(c) is particularly important because the Glu changes were measured 6 months before the T2-hyperintensity appeared and before the actual lesion volume could be observed. This correlation suggests that changes of Glu in WM including and surrounding the future lesions could also predict their volume. Together, these results underline the relevance of Glu to predict both the extent and severity of the subsequently forming lesions.

The relevance of increased WM Glu as biomarker of MS progression and clinical disability has been already demonstrated in one recent clinical study[7] where the ratio Glu/tNAA was associated with decline of brain volume and higher clinical disability.

No changes in tCr or Cho were found in the pre-lesional period, in constrast with Tartaglia et al.[8] This discrepancy could be explained by a difference in methodology. We investigated longitudinal absolute metabolite concentration changes, whereas Tartaglia et al. followed a cross-sectional approach. Additionally, the metabolite mask-matching analysis used in this study is beneficial for the precision of the results as the method corrects for intertwining of several tissue types (NAWM, GM, cerebrospinal fluid and lesional WM), and anatomical regions, known to differ naturally in terms of metabolite concentrations.[32]

Lesion formation period

In the lesion formation period, the lesions showing a high increase in tCr exhibited a small increase in AD and RD and vice versa. Likewise, when a high increase of Cho was observed, the increase in RD remained small. In fact, splitting the lesions into two groups ("mild" and "severe") based on the RD increase demonstrated that the effect is driven by the "mild" lesions: increases of tCr and Cho were only present in the "mild" lesion group. The increase of RD, which might reflect demyelination,[33] is a strong predictor for evolution of the new lesion into a black hole (a sign of permanent and severe tissue damage).[22,23] The metabolite variation in time and strong heterogeneity between lesions has already been described in magnetic resonance spectroscopy studies of acute WM lesions[9–12] but the origin of this variability remained unclear.

The observed increases of tCr and Cho could potentially be interpreted as a sign of remyelination, limiting damage in new lesions. This role for tCr was suggested before by a study that followed new contrast-enhancing lesions over time. These lesions showed increases of tCr, which was interpreted as a reflection of repopulation of the lesions with oligodendrocytes and, hence, remyelination.[13]

Our findings suggest that early metabolite changes predict the appearance and also the severity of newly forming lesions. Monitoring these changes could be useful for choice and timing of pharmacotherapeutical interventions, but also for understanding disease heterogeneity and clinical progression. Especially as the increasing Glu before lesion appearance could also be interpreted as an early leakage of the blood brain barrier causing inflammatory infiltrates stimulating clustering of activated glial cells.

Methodological considerations and future directions

Larger sample sizes, perhaps with multiple MRSI slabs, applied and measured, may be needed to support our results further. Furthermore, the actual time of lesion formation could not be determined more precisely in this study than with the 6-month window of the lesion formation period. However, this should not affect the validity of our results. Indeed, DTI metrics changes recorded in the WM lesions of MS patients were reported to appear during the first month[23] or 2 months[34] after enhancement, which falls entirely within the 6 months of the formation period taken into account here. Only the few lesions occurring in the last month of the formation period could have a possible incomplete increase of AD and RD that might have slightly reduced the overall effect.

It is important to mention that changes in DTI metrics and metabolite concentrations are measured on different mask sizes and that the lesion volume could be related to the results. Indeed, due to the dilation, the actual relative content of T2-hyperintense tissue in MRSI-LMs of small lesions is lower than for larger lesions. Therefore, it is important to consider this effect when interpreting correlation between metabolite changes in MRSI-LM and DTI metrics changes.

Imaging more frequently between time points and including contrast-enhancement could provide a more accurate characterization of metabolite changes as these are known to fluctuate in the month following the lesion formation.[12,13] Including a T1-weighted spin-echo sequence would enable the correlation of our findings to chronic T1-hypointensities, as a clinically used measure of (severe and/or lasting) tissue destruction.[35] Future studies are also required to observe whether a decrease of tNAA follows eventually at longer follow-up, and how this would be related to our findings.

Conclusion

Our results show that a Glu increase precedes the appearance and predicts the volume of the new T2 lesions in the MS WM, and that tCr and Cho increases predict the severity of tissue damage within those lesions. These findings are of essence for understanding lesion development in MS, and for therapeutic strategies or disease prediction.

Acknowledgements

A.M.K. and O.T.W. contributed equally to this work.

Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The author(s) declared receipt of the following financial support for the research, authorship, and/or publication of this article: This work was sponsored by the Canadian MS Society, the Dutch MS Research Foundation, grant number 09-358d and by Biogen Idec. Biogen Idec did not participate in any aspect of the design, performance and manuscript editing (including data collection, data management, data analysis and interpretation, or preparation) of this investigator initiated study. Final approval for the work and article was given by the authors.

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