On cerebral lupus: from pathogenesis to clinical outcomes

Magro Checa, C.

6

CHANGES IN WHITE MATTER MICROSTRUCTURE SUGGEST AN INFLAMMATORY ORIGIN OF NEUROPSYCHIATRIC SYSTEMIC LUPUS ERYTHEMATOSUS

César Magro-Checa, Ece Ercan, Ron Wolterbeek,
Bart Emmer, Nic J. van der Wee, Huub A. Middelkoop,
Nyika D. Kruyt, Itamar Ronen, Mark A. van Buchem,
Tom W.J. Huizinga, Gerda M. Steup-Beekman

Arthritis Rheumatol. 2016 Aug;68(8):1945-54.

ABSTRACT

Objective: To assess white matter (WM) and gray matter (GM) magnetization transfer ratio histogram peak heights (MTR-HPHs) in different subsets of patients with neuropsychiatric systemic lupus erythematosus (NP-SLE) who have unremarkable findings on 3T magnetic resonance imaging of the brain and to evaluate whether these values could be used to highlight different clinically suspected underlying pathogenic processes or identify the clinical NP-SLE status or whether they could be associated with a specific NP-SLE syndrome.

Methods: Sixty-four SLE patients with neuropsychiatric symptoms were included. The initial NP-SLE diagnosis and suspected underlying pathogenic process were established by multidisciplinary evaluation. The final diagnosis was made after also considering the disease course 6–18 months later. Thirty-three patients with central nervous system (CNS) NP-SLE and 31 SLE patients with neuropsychiatric symptoms unrelated to SLE (non–SLE-related NP) were included. Twenty SLE patients without neuropsychiatric symptoms and 36 healthy control subjects were included for comparison. Differences in the WM and GM mean MTR-HPHs and between the different NP-SLE subgroups (CNS NP-SLE diagnosis, NP-SLE phenotype [inflammatory or ischemic], and clinical changes after treatment) and the relationship to NP-SLE syndromes were evaluated.

Results: Patients with inflammatory NP-SLE had significantly lower WM MTR-HPHs than did the healthy controls, the SLE patients, and the non–SLE-related NP patients. Cognitive disorder, mood disorder and psychosis were related to lower WM MTR-HPH values and cerebrovascular symptoms to higher values. Furthermore, the mean MTR-HPHs in the WM increased when the clinical status of the NP-SLE patients improved.

Conclusion: Measurement of MTR-HPH of the WM has the potential to identify inflammatory NP-SLE with CNS involvement. This finding underscores the usefulness of this technique for the detection of cerebral changes in NP-SLE patients and for the assessment of clinical changes after treatment.

Systemic lupus erythematosus (SLE) is an autoimmune disease characterized by acute or chronic inflammation of multiple organs (1). Nervous system involvement in SLE, which is referred to as neuropsychiatric SLE (NP-SLE), leads to a broad, nonspecific, and heterogeneous group of NP manifestations (1,2). In 1999, the American College of Rheumatology (ACR) published a consensus document describing the diagnostic and exclusion criteria for 19 NP-SLE syndromes (3). Although widely used, its effectiveness is limited and NP-SLE remains a diagnosis per exclusion. Thus, in clinical practice, clinical suspicion of a certain pathogenic process underlying the clinical symptoms drives the therapeutic choice in these patients (4–6).

Two main underlying pathophysiologic processes have been described in NP-SLE, based on pathologic changes in humans and on findings in animal models. The inflammatory process (inflammatory NP-SLE) has been associated with dysfunction due to pathogenic antibodies and a disrupted blood–brain barrier, while the thrombotic process (ischemic NP-SLE) has been associated with focal neurologic deficits that can be attributed to interruption of blood flow in a specific brain region (5–7). Consistent with the suspected mechanism, therapy will be directed at the inflammation, with the use of immunosuppressive medications, or at the ischemia, with the use of antiaggregant and/or anticoagulant medications. These two phenotypes can also coexist.

So far, both the characterization of a certain NP-SLE phenotype and the correct attribution of NP events to SLE or to an alternative cause remain a challenge (8). None of the diagnostic tests currently used in clinical practice is specific for any NP-SLE manifestation or phenotype. Although magnetic resonance imaging (MRI) is the neuroimaging technique of choice in NP-SLE, this technique yields unremarkable findings in a significant proportion of patients, independently of the NP-SLE syndrome and its severity (8,9). There is thus an imperative need for radiologic techniques that help in the diagnostic process of NP-SLE and in the identification of NP-SLE phenotypes (2).

Magnetization transfer imaging (MTI) is a quantitative MRI technique known to be useful in the detection of cerebral abnormalities in brain tissue that looks normal on conventional MRI. This technique is based on the application of off-resonance radiofrequency pulses. Measurement of signal intensity with and without the application of these pulses allows the calculation of an index called the magnetization transfer ratio (MTR), which indirectly reflects the integrity of macromolecular structures (e.g., myelin) that exchange magnetization with the surrounding water (10,11). Among all of the MTI parameters, the histogram peak height (HPH), or the proportion of brain pixels at the most common MTR value, is the most informative parameter in NP-SLE without explanatory MRI findings. These values have been used as a quantitative estimate of tissue microstructural integrity in NP-SLE (12,13).

In preliminary investigations, Bosma and co-workers (14,15) observed a significantly lower whole-brain MTR-HPH in both active and past NP-SLE when compared with healthy controls. Those authors found an association between MTR-HPH and neurocognitive impairment and suggested that neuronal dysfunction may underlie central nervous system (CNS) involvement in NP-SLE (16). It has also been demonstrated that SLE patients with a history of NP had markedly lower gray matter (GM) MTR-HPHs than did healthy controls (17). Emmer and coworkers (18) showed how decreased whole-brain MTR-HPHs in patients with active NP-SLE increased when the clinical status improved, underscoring the possible partial reversibility of the previously observed abnormalities. Those authors also showed that in NP-SLE, there is a relationship between MTR-HPHs and neuronal impairment, as revealed by other quantitative neuroimaging techniques, such as diffusion-weighted imaging and proton magnetic resonance spectroscopy (13,19).

Despite these promising data, MTI has been applied only in a limited number of patients. The above-mentioned findings have never been reproduced in a NP-SLE cohort assessed through a multidisciplinary approach and followed prospectively. Prospective follow-up is essential for a diagnosis of NP-SLE. In the acute clinical setting, recognizing the cause of NP-SLE can be difficult, whereas at follow-up, the diagnosis can be assessed more reliably since the clinical course and response or failure to treatment provide diagnostic information.

The purposes of our study were to assess white matter (WM) and GM MTR-HPHs in a well-defined, prospectively followed cohort of SLE patients with NP symptoms that were either related or unrelated to SLE, to investigate whether these parameters may highlight different pathogenic NP-SLE processes (inflammatory or ischemic), and to reproduce previous findings published by our group in an evaluation of whether these parameters indicate the clinical NP-SLE status before and after treatment and whether they are related to different NP-SLE syndromes.

PATIENTS AND METHODS

Data source and population

All patients were admitted for a 1-day period to the Leiden University Medical Center. Our hospital serves as a national referral center for NP-SLE in the Netherlands. From September 1, 2007 through March 31, 2012, a total of 183 patients suspected of having NP involvement due to SLE were evaluated in the Leiden NP-SLE clinic. All patients underwent a standardized multidisciplinary medical examination, as well as extensive neuropsychological testing, serologic assessment, and brain MRI. Patients were classified according to the ACR 1982 revised criteria for SLE (20,21). SLE disease activity was determined with the use of the

Systemic Lupus Erythematosus Disease Activity Index 2000 (SLEDAI-2K) (22). Irreversible damage due to SLE was assessed with the Systemic Lupus International Collaborating Clinics (SLICC)/ACR damage index (SDI) (23). The SLEDAI-2K and SDI values were calculated both with and without NP manifestations. Soon after evaluation, a consensus meeting took place. Further descriptions of the multidisciplinary evaluation and laboratory examination are available elsewhere (6,24). All patients were closely monitored by the referring physician and reevaluated by our group 6–18 months after the first visit. Twenty SLE patients without NP symptoms and 36 age-matched healthy control subjects were also included in this study. Patients over the age of 70 years were excluded. Written informed consent was obtained from all patients. The study was approved by the local medical ethics committee and was carried out in compliance with the Declaration of Helsinki.

NP-SLE subgroups

Diagnosis of NP-SLE was made by multidisciplinary consensus, and NP diagnoses were classified according to the ACR 1999 definitions of NP-SLE (3,20,21). More than 1 NP diagnosis per patient was possible. We included in the NP-SLE group only patients with at least 1 NP-SLE syndrome involving the CNS. For each NP-SLE patient, a suspected pathogenic mechanism was also assessed. We differentiated between inflammatory and ischemic NP-SLE, as discussed above. Both inflammatory and ischemic phenotypes could coexist in the same patient. Changes in the clinical NP status between the first and second visits were assessed 6–18 months later and were classified as worse, stable, or improved by multidisciplinary consensus (rheumatology [C-MC, TWH, and GMS-B], neurology [NDK], psychiatry [NJvdW], neuropsychology [HAM], and neuroimaging [BE and MAvB]). In an important subgroup of SLE patients, the NP symptoms were explained by another diagnosis. These SLE patients with NP symptoms unrelated to SLE (non–SLE-related NP) were considered a different subgroup. During follow-up, none of the patients in the 2 groups with NP symptoms (n = 64) developed new NP symptoms.

MRI protocol and scoring

All patients underwent brain MRI following the same protocol and using the same scanner on a regular course of maintenance. All scans were performed on a 3-Tesla MRI scanner (Achieva, Philips Healthcare, Best, The Netherlands). The protocol included high-resolution T_1-weighted, T_2-weighted and fluid-attenuated inversion recovery (FLAIR) sequences, followed by a T_1-weighted sequence after intravenous administration of gadolinium contrast agent. An experienced radiologist (BE), who was blinded to the clinical status of the patients, examined visually all MRIs for the presence of abnormalities and for its suitability for MTI. To avoid the influence of ischemic areas due to thromboembolic processes on our results, we excluded patients with radiological evidence of other than incidental small (> 5mm)

infarctions and moderate atrophy measured by Pasquier scale (grade > 2; widened sulci, volume loss of the gyri). This scale, the most used visual rating scale (scores 0-3) for cortical atrophy, considers the volume of the gyri and width of the sulci (25). Subsequently, the differential diagnosis of ischemic NP-SLE without macroscopic MRI abnormalities included still cerebrovascular disease but also demyelinating syndromes and complex migraines.

MTI protocol

MTI-scans were performed using the same acquisition parameters for all NP-SLE, NP-non-SLE and SLE patients and HC. MTR data were obtained by using a 3-dimensional gradient echo sequence with an echo repetition/time of 100/11 msec and a low flip angle of 9°, to achieve minimal T_1-weighting. Twenty slices of 7.2mm thickness were acquired in an axial orientation, with a field of view = 224 × 180 × 144 mm^3 and acquisition matrix = 224 × 210 (voxel size 0.875 × 0.875 mm^2). To reduce acquisition time, segmented Echo-Planar Imaging (EPI) was applied, with 13 k-space profiles collected per excitation pulse (EPI factor 13). Two consecutive sets of axial images were acquired. The first set was performed in combination with a radiofrequency saturation pulse and the second without. Total scanning time was 1 minute and 8.3 seconds.

Image processing

For post-processing of magnetization transfer images, all images were transferred to an offline Linux workstation. All MTR processing steps were performed using software from the Oxford University Centre for Functional Magnetic Resonance Imaging of the Brain (FMRIB) software library (FSL) (26). MTR was defined as flows:

$$\mathbf{MTR = ([M_0 - M_S]/M_0) \times 100}$$

where M_s represents the signal intensity of voxels with saturation; and M_0, the signal intensity of voxels without saturation. Skull stripping was performed using FSL BET (27). A detailed description of the segmentation process based on T_1-weighted image and the way in which the resulting tissue masks were applied to the original MTR maps to calculate the tissue MTR maps (WM and GM) has been previously reported (13). To avoid the partial-volume effect of cerebrospinal fluid (CSF) at the tissue borders, the resulting maps were eroded in plane. From the remaining voxels, only those for which the probability of belonging to WM > 85% and GM > 80% were considered for the histogram analysis. All parenchyma segmentation was based on hard binary segmentations of GM and WM. All images were inspected visually to confirm adequate extraction of intracranial contents.

MTR histogram analysis

From the MTR maps, WM and GM MTR histograms were created with 100 bins and a bin size of 1. The first bin was excluded since it contains the voxels with an intensity of zero. The remaining 99 bins were taken into account for the subsequent calculations. MTR histograms were normalized for intracranial volume by dividing the number of voxels for each MTR value by the total number of CSF, WM and GM voxels. The corresponding peak height (PH) and peak location (PL) were calculated for WM and GM based on each normalized histogram using an in-house Matlab ® code. PL is an indicator of which MTR value is occurring more often. PH is a measure of the voxels fraction found to have the MTR value of the peak location. None of the WM or GM HPHs were used for clinical considerations.

Statistical analysis

The statistics included as primary dependent measures were the HPHs from the segmented WM and GM. Both were normally distributed. Equality of variances in WM and GM HPHs between NP-SLE, NP-non-SLE, SLE patients and controls was assessed using Levene's test. Between-group differences on WM and GM HPHs were evaluated using one-way-ANOVA´s (pairwise comparisons). In the events of unequal variances, appropriate adjustments according to Tamhane's procedure in the pairwise comparisons of the means were performed. Analysis of covariance was performed to analyze the influence of disease duration, SLEDAI-2K, SDI, smoking status, hypertension and anticardiolipin antibodies (aCL) on the differences on mean PH values between groups. The association between NP-SLE syndromes and HPHs values was assessed by independent T-test analysis in every NP-SLE syndrome present in > 5 patients taking into account a possible inequality of variances. Paired-samples **t**-test was performed to test for significant mean HPHs differences before and after treatment of active NP-SLE patients. Statistical analysis was performed with SPSS version 20.0 for Windows (IBM SPSS statistics, Chicago, IL, USA).

RESULTS

Patient selection and characterization

From all evaluated patients, 135 (73.8%) fulfilled the revised ACR criteria for SLE. In 59 patients (43,7%) of these patients a diagnosis of CNS NP-SLE was established in the second visit, whereas in the remaining patients the NP complaints were not directly attributed to SLE. After MRI evaluation, a total of 33 patients with CNS NP-SLE and 31 NP-non-SLE patients were suitable for our MTR study. The rest of the patients were excluded due to the presence of abnormalities in the conventional MRI. **Table 1** shows clinical characteristics and autoantibody profiles of the study subjects at the time of the first MRI. SLEDAI-2K with and without NP symptoms and SDI with NP symptoms were significantly higher in the NP-

SLE group. No differences were found for SDI without NP symptoms. Among the patients diagnosed with CNS NP-SLE, 22 were diagnosed with inflammatory NP-SLE and 11 with ischemic NP-SLE. Fifty-four different ACR NP syndromes were established.

White and grey matter MTR peak heights and NP-SLE diagnosis

The mean and standard deviation (SD) of the WM and GM MTR-HPHs and the mean differences between the study groups are respectively summarized in **Table 3** and **Table 4**. NP-SLE patients with CNS involvement had significantly lower WM MTR-HPH than HC ($P < 0.001$) and SLE patients ($P = 0.001$). No differences were found between NP-SLE and NP-non-SLE ($P = 0.114$). NP-non-SLE had significantly lower WM MTR-HPH than HC ($P < 0.001$). After adjustment with Tamhane's procedure no statistically differences were found between NP-non-SLE and SLE patients ($P = 0.063$). Furthermore, no statistically significant differences were found for WM when SLE and HC were compared.

We did not find any mean GM MTR-HPH difference between the subgroups. Control for differences attributable to disease duration, SLEDAI-2K, SDI, smoking status, hypertension and aCL did not reveal any significant influence on previous calculations. **Figure 1** shows the mean WM MTR histograms after correction for intracranial volume for all the NP-SLE, NP-non-SLE, SLE patients and HC.

Table 1 Characteristics of the study subjects

	NPSLE (n = 33)	NP-non-SLE * (n = 31)	SLE ** (n = 20)	Healthy controls (n = 36)
Age, mean ± SD years	37.2 ± 13.3	39.4 ± 14.9	41.1 ± 11.1	40.1 ± 11.8
Sex, no. female/male	29/4	28/3	18/2	32/4
SLE disease duration, mean ± SD years	5.2 ± 5.9	7.2 ± 7.3	8.8 ± 5.9	–
Neuropsychiatric symptoms duration, mean ± SD years	1.2 ± 2.7	2.7 ± 3.3	–	–
SLEDAI-2K without NP	6.8 ± 4.4	4.3 ± 3.2[b]	2.7 ± 2.4 [a]	–
SLEDAI-2K with NP	13.6 ± 5	4.3 ± 3.2 [a]	2.7 ± 2.4 [a]	–
SDI without NP	1.4 ± 1.2	1 ± 1.1	1.2 ± 1.2	–
SDI with NP	2.2 ± 1.4	1.2 ± 1.1 [a]	1.2 ± 1.2 [b]	–
ACR 1982 criteria for SLE †				
Malar Rash	16 (48.5)	14 (45.2)	11 (55)	–
Discoid rash	2 (6.1)	6 (19.4)	5 (25)	–
Photosensitivity	10 (30.3)	15 (48.4)	11 (55)	–
Mucosal ulcers	8 (24.2)	9 (29)	12 (60)	–
Arthritis	25 (75.7)	20 (64.5)	18 (90)	–
Serositis	9 (27,3)	10 (32.2)	3 (15)	–
Renal disorder	9 (27.3)	9 (29)	4 (20)	–
Neurological disorder	13 (39,4)	0 (0)	0 (0)	
Hematologic disorder	17 (51.5)	14 (45.2)	15 (75)	–
Immunologic disorder	29 (87.9)	21 (67.7)	18 (90)	–
Positive ANA	31 (93.9)	30 (96.8)	20 (100)	–
Autoantibodies and complement †				
aCL IgG	8 (24.2)	5 (16.1)	2 (10)	–
aCL IgM	1 (3)	2 (6.5)	2 (10)	–
LAC	13 (39.4)	5 (16.1)	3 (15)	–
Antinuclear antibody	29 (87.9)	24 (77.4)	18 (90)	–
Anti-dsDNA	13 (39.4)	9 (29)	9 (45)	–
ENA	16 (48.5)	16 (51.6)	8 (40)	–
Anti-SSA	9 (27.3)	11 (35.5)	6 (30)	–
Anti-SSB	3 (9.1)	6 (19.4)	2 (10)	–
Anti-RNP	8 (24.2)	3 (9.7)	4 (20)	–
Anti-Sm	6 (18.2)	3 (9.7)	4 (20)	–
C1q low	3 (9.1)	1 (3.2)	1 (5)	–
C3 low	13 (39.4)	10 (32.3)	3 (15)	–
C4 low	12 (36.4)	6 (19.4)	5 (25)	–

aCL: anticardiolipin antibodies; ACR: American College of Rheumatology; ANA: antinuclear antibody; LAC: Lupus anticoagulant; NP: neuropsychiatric symptoms; NPSLE: neuropsychiatric systemic lupus erythematosus; SLE: systemic lupus erythematosus; SDI: systemic lupus international collaborating clinics (SLICC)/American College of Rheumatology damage index; SLEDAI-2K: Systemic Lupus Erythematosus Disease Activity Index 2000.
* SLE patients with NP complaints non associated with CNS involvement due to SLE
** SLE patients without NP complaints
† Number and percentage per group
a. $P < 0.001$ when compared with NPSLE
b. $P < 0.05$ when compared with NPSLE

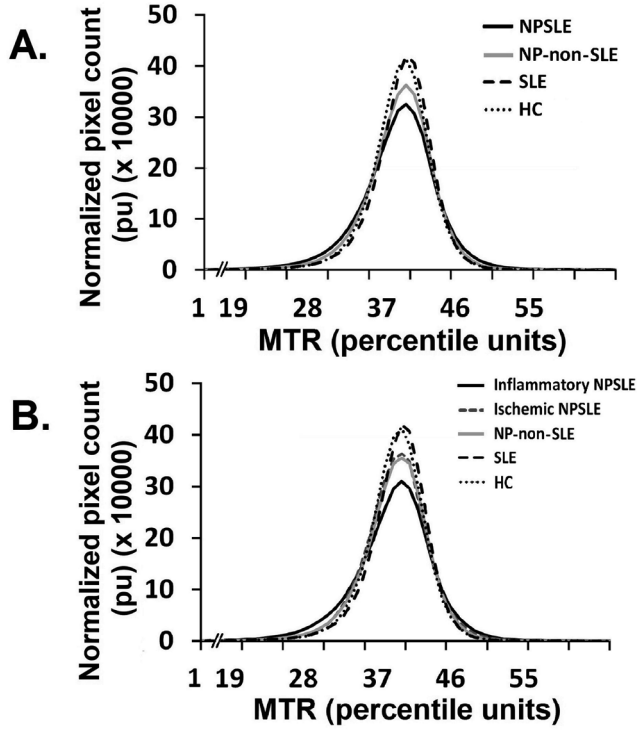

Figure 1. Average white matter magnetization transfer ratio (MTR) histograms. Mean MTR histograms after correction for intracranial volume are shown in A, patients with neuropsychiatric systemic lupus erythematosus (NPSLE), patients with NP symptoms unrelated to the underlying SLE (non–SLE-related NP), SLE patients without NP symptoms, and healthy control (HC) subjects, as well as in B, patients with inflammatory NPSLE, ischemic NPSLE, non–SLE-related NP, SLE patients without NP symptoms, and healthy control subjects. pu = percentage units.

White and grey matter MTR peak heights and NP-SLE phenotypes

The mean and SD of the WM and GM MTR-HPHs are presented in **Table 3.** The mean differences between the study groups are presented in **Table 4**. Patients with inflammatory NP-SLE had significantly lower WM MTR-HPH when compared with HC (WM $P < 0.001$), SLE (WM $P < 0.001$) and NP-non-SLE (WM $P = 0.023$). Moreover, inflammatory NP-SLE had a significantly lower WM MTR-HPH when compared with ischemic NP-SLE ($P = 0.001$). No statistically significant differences were found for WM when we compared ischemic NP-SLE with HC, NP-non-SLE or SLE. Inflammatory NP-SLE had also significantly lower GM MTR-HPH when compared with SLE ($P = 0.044$) but we did not find other differences when compared with other subgroups. We did not find any statistically significant difference for GM when ischemic NP-SLE patients were compared with HC, NP-non-SLE and SLE. Control for differences attributable to disease duration, SLEDAI-2K, SDI, smoking status, hypertension and aCL did not reveal any significant influence on previous calculations. WM MTR histograms in the 5 study groups are shown in **Figure 1**.

Table 2. Comparison of white matter and grey matter MTR-HPHs in the study groups *

		WM MTR-HPH §	GM MTR-HPH §
Healthy controls	36	43.37 ± 5.11	10.01 ± 2.51
SLE †	20	42.74 ± 6.22	10.02 ± 1.92
NP-non-SLE ‡	31	38.35 ± 4.64	9.81 ± 3.68
NPSLE	33	34.62 ± 7.55	8.56 ± 3.31
Phenotype			
Inflammatory NPSLE	22	32.22 ± 7.76	7.71 ± 3.25
Ischemic NPSLE	11	39.42 ± 4.21	10.25 ± 2.85

CNS: central nervous system; GM: grey matter; MTR-HPH: magnetization transfer ratio histogram peak height; NPSLE: neuropsychiatric SLE; SLE: systemic lupus erythematosus; WM: white matter.
* Values are the mean ± standard deviation.
† SLE patients without neuropsychiatric complaints
‡ SLE patients with neuropsychiatric complaints non associated with CNS involvement due to SLE
§ Peak height values were multiplied by 10,000 for readability

Table 3. Mean differences after Tamhane procedure of the WM and GM MTR-HPHs between the study groups *

	WM Peak height	GM Peak height
NPSLE diagnosis		
NPSLE – Healthy controls	−8.74 (0.000) † [−13.02 to −4.47]	−1.45 (0.247) [−3.39 to 0.48]
NPSLE – SLE	−8.12 (0.001) § [−13.38 to −2.85]	−1.64 (0.150) [−3.61 to 0.32]
NPSLE – NP-non-SLE	−3.73 (0.114) [−7.98 to 0.51]	−1.24 (0.654) [−3.63 to 1.14]
NP-non-SLE – Healthy controls	−5.01 (0.000) † [−8.24 to −1.77]	−0.21 (1.000) [−2.35 to 1.93]
NP-non-SLE – SLE	−4.39 (0.063) [−8.93 to 0.16]	−0.39 (0.997) [−2.56 to 1.76]
SLE – Healthy controls	−0.62 (0.999) [−5.19 to 3.94]	0.19 (1.000) [−1.45 to 1.84]
NPSLE phenotype		
NPSLE inflammatory – Healthy controls	−11.14 (0.000) † [−16.74 to −5.54]	−2.29 (0.073) [−4.71 to 0.12]
NPSLE inflammatory – SLE	−10.52 (0.000) † [−16.93 to −4.11]	−2.48 (0.044) § [−4.93 to −0.04]
NPSLE inflammatory – NP-non-SLE	−6.13 (0.023) § [−11.71 to −0.55]	−2.09 (0.296) [−4.91 to 0.72]
NPSLE inflammatory – NPSLE ischemic	−7.19 (0.001) § [−11.36 to −3.02]	−2.53 (0.276) [−5.96 to 0.89]
NPSLE ischemic – Healthy controls	−3.94 (0.165) [−8.75 to 0.86]	0.24 (1.000) [−2.89 to 3.37]
NPSLE ischemic – SLE	−3.32 (0.607) [−9.05 to 2.41]	0.47 (1.000) [−3.11 to 3.19]
NPSLE ischemic – NP-non-SLE	1.06 (0.999) [−3.73 to 5.86]	0.44 (1.000) [−2.92 to 3.81]

GM: grey matter; HC: healthy controls; MTR-HPH: magnetization transfer ratio histogram peak height; NPSLE: neuropsychiatric SLE; SLE: systemic lupus erythematosus; NP-non-SLE: systemic lupus erythematosus with neuropsychiatric complaints non-SLE related; WM: white matter.
* Mean difference (P-value) [95% CI]
† Indicates significance level at $P < 0.001$
§ Indicates significance level at $P < 0.05$

White and grey matter MTR peak heights and NP-SLE syndromes

Independent T-test analysis was performed for every NP-SLE syndrome present in > 5 patients. Patients with cerebrovascular disease (n = 11), psychosis (n = 8), headache (n = 8), seizure (n = 5), cognitive disorder (n = 9) and mood disorder (n = 10) were analyzed individually. Psychosis was associated with a lower WM MTR-HPH ($P = 0.033$) and GM MTR-HPH ($P = 0.029$). We also found an association between a lower WM MTR-HPH and cognitive disorder ($P = 0.047$) and mood disorder ($P = 0.025$). We did not find any association between GM MTR-HPHs and cognitive disorder or mood disorder. Furthermore, cerebrovascular disease was associated with higher WM MTR-HPH ($P = 0.006$). No associations were found between MTR-HPHs and patients with headache or seizure.

White matter MTR peak heights and clinical changes

From all the twenty NP-SLE patients considered to have active CNS disease during the first visit, eleven patients improved after treatment, seven were classified as stable and 2 patients deteriorated. Mean histogram and standard deviation WM MTR-HPH of all patients at first visit was 31.51 ± 7.83. On the follow-up visit these values increased and mean MTR-HPH was 39.07 ± 6.56. WM MTR histograms after correction for intracranial volume before and after treatment are shown in **Figure 2**. In all NP-SLE patients that clinically improved, the mean WM MTR-HPH increased in 9.81 ± 5.94 [range 5.81 to 13.81] ($P < 0.000$). WM MTR-HPH mean difference of patients classified as stable on the second visit were 2.48 ± 4.65 [range 1.81 to 6.79] ($P = 0.207$). In the two patients who deteriorated, a decrease on the WM MTR-HPH between the first and second MRI was observed −10.32 ± 0.41 [range −14.01 to −6.63] ($P = 0.018$).

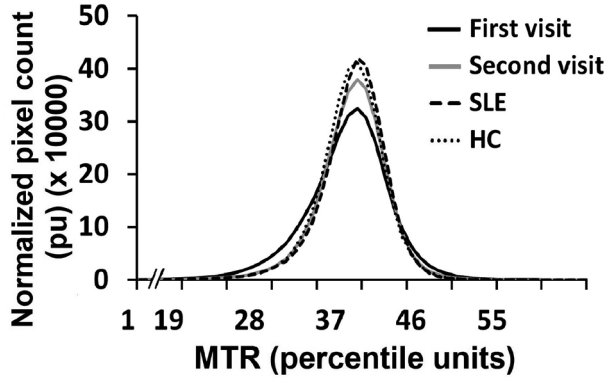

Figure 2. Average magnetization transfer ratio histograms of white matter from active NPSLE patients on the first visit and after treatment on the second visit. SLE patients without neuropsychiatric complaints and healthy controls (HC) on the basal visit are also included for comparison.

DISCUSSION

This study is the first to show that NP-SLE patients with an inflammatory phenotype have significantly lower WM MTR-HPHs than do ischemic NP-SLE, non–SLE-related NP, or SLE patients or healthy controls. We also found that WM MTR-HPH is sensitive to clinical changes. Based on these findings, we propose that the WM MTR-HPH is a potentially valuable tool for use in the diagnosis and follow-up of inflammatory NP-SLE.

Inflammatory NP-SLE is thought to reflect neuronal dysfunction mediated by inflammatory factors, autoantibodies, and increased SLE disease activity. Apart from global and localized ischemic changes, histopathologic data in NP-SLE show parenchymal edema, glial hyperplasia, and diffuse neuronal/axonal loss (7). It has been hypothesized that MTR changes are associated with all of these findings and may thus also explain our results (13,18). In multiple sclerosis, MTR abnormalities have been described as a useful tool for assessing disease burden and evaluating disease progression (28). However, demyelination is not a primary phenomenon in NP-SLE, and other mechanisms may play a more important role in these MTR changes (8). The fact that the WM MTR-HPHs in patients with ischemic NP-SLE, mainly seen in those with cerebrovascular symptoms, were lower than those in the healthy controls and significantly higher than those in patients with inflammatory NP-SLE may suggest cumulative chronic damage of the brain, as reported previously (13,16). Furthermore, mean MTR-HPHs at the second visit were, on average, closer to those in ischemic NP-SLE patients, probably reflecting residual effects or WM-specific and irreversible changes in patients with past inflammatory NP-SLE.

To our knowledge, this is the first study in which prospective follow-up was performed in order to avoid misclassification of the putative cause of NP symptoms in SLE. This standardized assessment is the most appropriate reference standard for diagnosis so far (29). In addition, we were able to include patients with CNS involvement without remarkable abnormalities on MRI. These well-defined data are an additional benefit of our study. This study also reproduced some data previously published by our group.

We found that NP-SLE patients and non–SLE-related NP patients have, on average, significantly lower WM MTR-HPHs than do healthy controls. Furthermore, the WM MTR-HPHs in NP-SLE patients were significantly lower on average than those in SLE patients, but no differences were found between SLE patients and non–SLE-related NP patients. The usefulness of whole-brain parenchyma or segmented tissue MTR-HPHs for the differentiation of SLE patients with NP symptoms has previously been reported (13–15,17,30,31). Studies based on other quantitative radiologic techniques, such as proton magnetic resonance spectroscopy and diffusion tensor imaging, have demonstrated a loss of WM integrity in SLE

patients and non–SLE-related NP patients as compared with healthy controls (13,32–34) . Using MTI, we found no differences betw een SLE patients and healthy controls, which m ay suggest that each technique identifies different aspects of the microstructural changes in the brains of SLE and NP-SLE patients. As previously reported, no differences between NP-SLE patients and non–SLE-related NP patients were found, probably because the NP-SLE group included both ischemic and inflammatory NP-SLE subgroups (13).

There may be 2 possible explanations for the lower WM MTR-HPH values in the non–SLE-related NP patients. Despite multidisciplinary assessment, we still might have misclassified some NP-SLE patients as having non–SLE-related NP. Additionally, the non–SLE-related NP group included a broad spectrum of active neurologic and psychiatric disorders, which may have influenced the MTR results, as lower MTR values have been previously reported in patients with behavioral, psychotic, and neurodegenerative disorders (35–37).

Cognitive dysfunction was associated with lower WM MTR-HPHs, as previously observed in other studies (13,16). We also found an association between psychosis and lower WM and GM MTR-HPHs, as well as between mood disorder and WM MTR-HPHs. In contrast, cerebrovascular disease was related to higher WM MTR-HPHs, and no associations for headache or seizure were noted. Cognitive dysfunction, psychosis, and mood disorder may share a similar pathogenic pathway as compared with other syndromes. However, these results may be related to the prevalence of certain syndromes and their activity at the time of MRI as well as to the heterogeneity of NP-SLE. As mentioned above, nonspecific microstructural changes of the brain tissue as mea sured by MTR have been found in several brain regions in patients with cognitive impairment, psychosis, and mood disorder (35–37).

As demonstrated previously (18), we have seen how brain involvement in patients with active NP-SLE with unremarkable findings on MRI is partially reversible when measuring WM MTR-HPHs. These values decreased or increased in parallel with the clinical status of the patients, as assessed by our multidisciplinary group. It has been suggested that these changes may be linked to the resolution or exacerbation of general inflammatory changes of the brain (7,18). It is unclear whether these MTR changes after treatment are associated with remyelination, as has been demonstrated in multiple sclerosis (18,38). Our data reinforce the idea that MTI, especially the MTR histogram analysis, may be a useful tool for evaluating disease progression and response to therapy.

Our results also show a lower GM MTR-HPH in patients with inflammatory NP-SLE as compared with those with SLE and a trend as compared with healthy controls. The difference between NP-SLE patients and healthy controls was previously reported by Steens and coworkers (17). The selective lowering of the GM MTR-HPH in patients with inflammatory

CNS NP-SLE without remarkable abnormalities on MRI may reveal GM-specific changes. However, these data should be viewed with caution, since several factors could affect these results. The presence of cortical atrophy, especially focal, has been observed in NP-SLE (8,9). Due to partial volume effects, the voxels analyzed in the parenchyma l cortex contain a mixture of GM, WM, and CSF. This may lead to a misclassification of those voxels as GM and, subsequently, to decreased GM MTR-HPHs. To avoid the effect of atrophy, we used the Pasquier scale for patient selection, as well as stringent thresholds for GM parenchyma analysis to reduce partial volume effects as much as possible without losing the representation of the segmented tissue type.

We were not able to reproduce other data previously published by our group in studies of a smaller number of patients. Steens and coworkers found an association between certain MTR values (WM and GM mean MTR and peak location) and positivity for IgM aCL, suggesting that these antibodies may be associated with diffuse brain involvement (17). This association between MTR values and aCL status was not further confirmed (13). We found no association between aCL and HPHs. Previously, an association between certain SLE criteria, such as arthritis and renal involvement, and MTR-HPHs was observed (13). In the present study, associations between HPHs and disease activity (SLEDAI-2K) were not found. We believe that our previous data may show false-positive associations based on the small sample size.

The main limitation of our study is the small number of patients per group and per syndrome. This is a generally recognized problem related to the low pre valence and the high heterogeneity of NP-SLE. We therefore cannot draw definite conclusions concerning the relationship between the MTR-HPH findings and NPS LE syndromes. Furthermore, due to matters of referral, some of the patients with inflammatory NP-SLE were evaluated in the NP-SLE clinic once they had started the immunosuppressive therapy. This may explain the higher variance in the NP-SLE group, and we believe that inflammatory NP-SLE would probably have shown lower values in comparison with other groups if none of these patients had received prior therapy.

A second limitation is that for research purposes, we selected patients with unremarkable findings on MRI, excluding a high proportion of patients to avoid the influence of thromboembolic processes. Our data can thus be extrapolated only to NP-SLE patients with unremarkable MRI findings, since the effect of the presence of infarcts and WM lesions on the MTR-HPHs values remains unknown. Another limitation of our study is the possible misclassification of inflammatory NP-SLE based on a good response to therapy, whereas the clinical response could have been the normal waxing and waning of the disease course or due to their inclusion in this group of nonspecific NP-SLE syndromes (headache, mood disorder, anxiety, and mild cognitive dysfunction). However, such misclassification would

lead to smaller differences between groups, and the real differences may therefore be even larger than we report here. A final limitation is that due to the impaired clinical status of some patients, we had to decrease the scanning time, which subsequently affected the resolution, resulting in partial volume effects, which may cause misclassification of GM and WM voxels.

In conclusion, this is the first study to demonstrate that WM MTR-HPHs might provide evidence of the presence of inflammatory NP-SLE. This study also confirmed the usefulness of this technique in the detection of cerebral changes in NP-SLE and in the assessment of clinical changes after treatment of patients with active disease. Moreover, a lower WM MTR-HPH was associated with cognitive dysfunction, mood disorder, and psychosis. Furth er studies are required to fully determine whether these data reflect the burden of SLE on the brain or whether they represent the severity of NP symptoms apart from the SLE. Our results are consistent with previous data reported by our group, thus broadening their significance. The findings of our study illustrate the value of MTR-HPH analysis as a potential radiologic biomarker that may help in the diagnostic process and follow-up of patients with NP-SLE and with the monitoring of future treatment trials.

REFERENCES

1. Arbuckle MR, McClain MT, Rubertone MV, Scofield RH, Dennis GJ, James JA, et al. Development of autoantibodies before the clinical onset of systemic lupus erythematosus. N Engl J Med 2003;349:1526–33.
2. Jeltsch-David H, Muller S. Neuropsychiatric systemic lupus erythematosus: pathogenesis and biomarkers. Nat Rev Neurol 2014;10:579–96.
3. ACR Ad Hoc Committee on Neuropsychiatric Lupus Nomenclature. The American College of Rheumatology nomenclature and case definitions for neuropsychiatric lupus syndromes. Arthritis Rheum 1999;42:599–608.
4. Hanly JG. ACR classification criteria for systemic lupus erythematosus: limitations and revisions to neuropsychiatric variables. Lupus 2004;13:861–4.
5. Hanly JG. Diagnosis and management of neuropsychiatric SLE. Nat Rev Rheumatol 2014;10:338–47.
6. Zirkzee EJ, Steup-Beekman GM, van der Mast RC, Bollen EL, van der Wee NJ, Baptist E, et al. Prospective study of clinical phenotypes in neuropsychiatric systemic lupus erythematosus; multidisciplinary approach to diagnosis and therapy. J Rheumatol 2012;39:2118–26.
7. Sibbitt WL Jr, Brooks WM, Kornfeld M, Hart BL, Bankhurst AD, Roldan CA. Magnetic resonance imaging and brain histopathology in neuropsychiatric systemic lupus erythematosus. Semin Arthritis Rheum 2010;40:32–52.
8. Sibbitt WL Jr, Sibbitt RR, Brooks WM. Neuroimaging in neuropsychiatric systemic lupus erythematosus [review]. Arthritis Rheum 1999;42:2026–38.
9. Luyendijk J, Steens SC, Ouwendijk WJ, Steup-Beekman GM, Bollen EL, van der Grond J, et al. Neuropsychiatric systemic lupus erythematosus: lessons learned from magnetic resonance imaging. Arthritis Rheum 2011;63:722–32.
10. Grossman RI, Gomori JM, Ramer KN, Lexa FJ, Schnall MD. Magnetization transfer: theory and clinical applications in neuroradiology. Radiographics 1994;14:279–90.
11. Price SJ, Tozer DJ, Gillard JH. Methodology of diffusion weighted, diffusion tensor and magnetisation transfer imaging. Br J Radiol 2011;84:S121–6.
12. Brooks WM, Jung RE, Ford CC, Greinel EJ, Sibbitt WL Jr. Relationship between neurometabolite derangement and neurocognitive dysfunction in systemic lupus erythematosus. J Rheumatol 1999;26:81–5.
13. Emmer BJ, Steup-Beekman GM, Steens SC, Huizinga TW, van Buchem MA, van der Grond J. Correlation of magnetization transfer ratio histogram parameters with neuropsychiatric systemic lupus erythematosus criteria and proton magnetic resonance spectroscopy: association of magnetization transfer ratio peak height with neuronal and cognitive dysfunction. Arthritis Rheum 2008;58:1451–7.
14. Bosma GP, Rood MJ, Huizinga TW, de Jong BA, Bollen EL, van Buchem MA. Detection of cerebral involvement in patients with active neuropsychiatric systemic lupus erythematosus by the use of volumetric magnetization transfer imaging. Arthritis Rheum 2000;43:2428–36.
15. Bosma GP, Rood MJ, Zwinderman AH, Huizinga TW, van Buchem MA. Evidence of central nervous system damage in patients with neuropsychiatric systemic lupus erythematosus, demonstrated by magnetization transfer imaging. Arthritis Rheum 2000;43:48–54.
16. Bosma GP, Middelkoop HA, Rood MJ, Bollen EL, Huizinga TW, van Buchem MA. Association of global brain damage and clinical functioning in neuropsychiatric systemic lupus erythematosus. Arthritis Rheum 2002;46:2665–72.
17. Steens SC, Admiraal-Behloul F, Bosma GP, Steup-Beekman GM, Olofsen H, le Cessie S, et al. Selective gray matter damage in neuropsychiatric lupus: a magnetization transfer imaging study. Arthritis Rheum 2004;50:2877–81.
18. Emmer BJ, Steens SC, Steup-Beekman GM, van der Grond J, Admiraal-Behloul F, Olofsen H, et al. Detection of change in CNS involvement in neuropsychiatric SLE: a magnetization transfer study. J Magn Reson Imaging 2006;24:812–6.
19. Bosma GP, Steens SC, Petropoulos H, Admiraal-Behloul F, van den Haak A, Doornbos J, et al. Multisequence magnetic resonance imaging study of neuropsychiatric systemic lupus erythematosus. Arthritis Rheum 2004;50:3195–202.
20. Hochberg MC, for the Diagnostic and Therapeutic Criteria Committee of the American College of Rheumatology. Updating the American College of Rheumatology revised criteria for the classification of systemic lupus erythematosus. Arthritis Rheum 1997;40:1725.
21. Tan EM, Cohen AS, Fries JF, Masi AT, McShane DJ, Rothfield NF, et al. The 1982 revised criteria for the classification of systemic lupus erythematosus. Arthritis Rheum 1982;25:1271–7.
22. Gladman DD, Ibanez D, Urowitz MB. Systemic Lupus Erythematosus Disease Activity Index 2000. J Rheumatol 2002;29:288–91.

23. Gladman DD, Urowitz MB, Goldsmith CH, Fortin P, Ginzler E, Gordon C, et al. The reliability of the Systemic Lupus International Collaborating Clinics/ American College of Rheumatology Damage Index in patients with systemic lupus erythematosus. Arthritis Rheum 1997;40:809–13.
24. Steens SC, Bosma GP, Steup-Beekman GM, le Cessie S, Huizinga TW, van Buchem MA. Association between microscopic brain damage as indicated by magnetization transfer imaging and anticardiolipin antibodies in neuropsychiatric lupus. Arthritis Res Ther 2006;8:R38.
25. Scheltens P, Pasquier F, Weerts JG, Barkhof F, Leys D. Qualitative assessment of cerebral atrophy on MRI: inter- and intraobserver reproducibility in dementia and normal aging. Eur Neurol 1997;37:95–9.
26. Smith SM, Jenkinson M, Woolrich MW, Beckmann CF, Behrens TE, Johansen-Berg H, et al. Advances in functional and structural MR image analysis and implementation as FSL. Neuroimage 2004;23 Suppl 1:S208–19.
27. Smith SM. Fast robust automated brain extraction. Hum Brain Mapp 2002;17:143–55.
28. Chen JT, Collins DL, Atkins HL, Freedman MS, Arnold DL. Magnetization transfer ratio evolution with demyelination and remyelination in multiple sclerosis lesions. Ann Neurol 2008;63:254–62.
29. Rutjes AW, Reitsma JB, Coomarasamy A, Khan KS, Bossuyt PM. Evaluation of diagnostic tests when there is no gold standard: a review of methods. Health Technol Assess 2007;11:iii, ix–51.
30. Dehmeshki J, Van Buchem MA, Bosma GP, Huizinga TW, Tofts PS. Systemic lupus erythematosus: diagnostic application of magnetization transfer ratio histograms in patients with neuropsychiatric symptoms—initial results. Radiology 2002;222:722–8.
31. Rovaris M, Viti B, Ciboddo G, Gerevini S, Capra R, Iannucci G, et al. Brain involvement in systemic immune mediated diseases: magnetic resonance and magnetization transfer imaging study. J Neurol Neurosurg Psychiatry 2000;68:170–7.
32. Appenzeller S, Li LM, Costallat LT, Cendes F. Evidence of reversible axonal dysfunction in systemic lupus erythematosus: a proton MRS study. Brain 2005;128:2933–40.
33. Schmidt-Wilcke T, Cagnoli P, Wang P, Schultz T, Lotz A, Mccune WJ, et al. Diminished white matter integrity in patients with systemic lupus erythematosus. Neuroimage Clin 2014;5:291–7.
34. Emmer BJ, Veer IM, Steup-Beekman GM, Huizinga TW, van der Grond J, van Buchem MA. Tract-based spatial statistics on diffusion tensor imaging in systemic lupus erythematosus reveals localized involvement of white matter tracts. Arthritis Rheum 2010;62:3716–21.
35. Kumar A, Gupta RC, Albert TM, Alger J, Wyckoff N, Hwang S. Biophysical changes in normal-appearing white matter and subcortical nuclei in late-life major depression detected using magnetization transfer. Psychiatry Res 2004;130:131–40.
36. Price G, Cercignani M, Chu EM, Barnes TR, Barker GJ, Joyce EM, et al. Brain pathology in first-episode psychosis: magnetization transfer imaging provides additional information to MRI measurements of volume loss. Neuroimage 2010;49:185–92.
37. Seiler S, Pirpamer L, Hofer E, Duering M, Jouvent E, Fazekas F, et al. Magnetization transfer ratio relates to cognitive impairment in normal elderly. Front Aging Neurosci 2014;6:263.
38. Schmierer K, Scaravilli F, Altmann DR, Barker GJ, Miller DH. Magnetization transfer ratio and myelin in postmortem multiple sclerosis brain. Ann Neurol 2004;56:407–15.

6