Changes in White Matter Microstructure Suggest an Inflammatory Origin of Neuropsychiatric Systemic Lupus Erythematosus

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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 (NPSLE) 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 NPSLE status or whether they could be associated with a specific NPSLE syndrome.

Methods. Sixty-four SLE patients with neuropsychiatric symptoms were included. The initial NPSLE 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) NPSLE 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 NPSLE subgroups (CNS NPSLE diagnosis, NPSLE phenotype [inflammatory or ischemic], and clinical changes after treatment) and the relationship to NPSLE syndromes were evaluated.

Results. Patients with inflammatory NPSLE 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 NPSLE patients improved.

Conclusion. Measurement of MTR-HPH of the WM has the potential to identify inflammatory NPSLE with CNS involvement. This finding underscores the usefulness of this technique for the detection of cerebral changes in NPSLE 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 (NPSLE), 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 NPSLE syndromes (3). Although widely used, its effectiveness is limited and NPSLE 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 NPSLE, based on pathologic changes in humans and on findings in animal models. The inflammatory process (inflammatory NPSLE) has been associated with dysfunction due to pathogenic antibodies and a disrupted blood–brain barrier, while the thrombotic process (ischemic NPSLE) 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 NPSLE 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 NPSLE manifestation or phenotype. Although magnetic resonance imaging (MRI) is the neuroimaging technique of choice in NPSLE, this technique yields unremarkable findings in a significant proportion of patients, independently of the NPSLE syndrome and its severity (8,9). There is thus an imperative need for radiologic techniques that help in the diagnostic process of NPSLE and in the identification of NPSLE 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 NPSLE without explanatory MRI findings. These values have been used as a quantitative estimate of tissue microstructural integrity in NPSLE (12,13).

In preliminary investigations, Bosma and coworkers (14,15) observed a significantly lower whole-brain MTR-HPH in both active and past NPSLE 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 NPSLE (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 NPSLE increased when the clinical status improved, underscoring the possible partial reversibility of the previously observed abnormalities. Those authors also showed that in NPSLE, 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 NPSLE cohort assessed through a multidisciplinary approach and followed prospectively. Prospective follow-up is essential for a diagnosis of NPSLE. In the acute clinical setting, recognizing the cause of NPSLE 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 NPSLE processes (inflammatory or ischemic), and to reproduce previous findings published by our group in an evaluation of whether these parameters indicate the clinical NPSLE status before and after treatment and whether they are related to different NPSLE 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 NPSLE 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 NPSLE clinic. All patients underwent a standardized multidisciplinary medical examination, as well as extensive neuropsychologic 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.

NPSLE subgroups. Diagnosis of NPSLE was made by multidisciplinary consensus, and NP diagnoses were classified according to the ACR 1999 definitions of NPSLE (3,20,21). More than 1 NP diagnosis per patient was possible. We included in the NPSLE group only patients with at least 1 NPSLE syndrome involving the CNS. For each NPSLE patient, a suspected pathogenic mechanism was also assessed. We differentiated between inflammatory and ischemic NPSLE, 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 [NJrdw], 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 according to the same protocol and using the same scanner. All scans were performed on a 3T MRI scanner (Achieva; Philips Healthcare). The protocol included high-resolution T1-weighted, T2-weighted, and fluid-attenuated inversion recovery sequences, followed by a T1-weighted sequence obtained after intravenous administration of gadolinium contrast agent. An experienced radiologist (BE) who was blinded with regard to the clinical status of the patients visually examined all MRIs for the presence of abnormalities and for their suitability for MTI. To avoid any influence of ischemic areas due to thromboembolic processes on our results, we excluded patients with radiologic evidence of other-than-incidental small (>5 mm) infarctions and moderate atrophy, as measured by the Pasquier scale (grade >2; widened sulci, volume loss of the gyri). This scale, the most frequently used visual rating scale for cortical atrophy (scored on a 0–3 scale), considers the volume of the gyri and width of the sulci (25). The differential diagnosis of ischemic NPSLE without macroscopic abnormalities on MRI included cerebrovascular disease as well as demyelinating syndromes and complex migraines.

MTI protocol. MTI scans were performed using the same acquisition parameters for all NPSLE, non–SLE-related NP, and SLE patients and healthy control subjects. MTR data were obtained by using a 3-dimensional gradient-echo sequence with a repetition time/echo time of 100/11 msec and a low flip angle of 9°, to achieve minimal T1 weighting. A total of 20 slices of 7.2 mm in thickness were acquired in an axial orientation, with a field of view of $224 \times 180 \times 144$ mm^3 and an acquisition matrix of 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 8.3 seconds.

Image processing. For postprocessing 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 follows:

$$MTR = ([M_0 - M_s]/M_0) \times 100$$

where M_0 represents the signal intensity of voxels without saturation, and M_s represents the signal intensity of voxels with saturation.

Skull stripping was performed using FSL BET software (27). A detailed description of the segmentation process based on the T1-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) is reported in detail elsewhere (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 data.

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 were calculated for WM and GM based on each normalized histogram using an in-house MatLab code. Peak location is an indicator of which MTR value is occurring more often. Peak height is a measure of the voxel fraction found to have the MTR value of the peak location. None of the WM or GM HPHs was used for clinical considerations.

Statistical analysis. Statistical analysis included as the primary dependent measures the HPHs from the segmented WM and GM. Both were normally distributed. Equality of variances in WM and GM HPHs between NPSLE patients, non–SLE-related NP patients, SLE patients, and healthy controls was assessed using Levene's test. Between-group differences in WM and GM HPHs were evaluated using one-way analysis of variance (pairwise comparisons). In the event of unequal variances, appropriate adjustments in the pairwise comparisons of the means were performed according to Tamhane's procedure. Analysis of covariance was performed to analyze the influence of disease duration, SLEDAI-2K, SDI, smoking status, hypertension, and anticardiolipin antibodies (aCL) on the between-group differences in mean peak height

Table 1.   Characteristics of the study patients, by diagnostic group*

	NPSLE patients (n = 33)	Non–SLE-related NP patients (n = 31)	SLE patients (n = 20)
Age, mean ± SD years	37.2 ± 13.3	39.4 ± 14.9	41.1 ± 11.1
Sex, no. female/male	29/4	28/3	18/2
SLE disease duration, mean ± SD years	5.2 ± 5.9	7.2 ± 7.3	8.8 ± 5.9
NP symptom duration, mean ± SD years	1.2 ± 2.7	2.7 ± 3.3	–
SLEDAI-2K, mean ± SD			
Without NP symptoms	6.8 ± 4.4	4.3 ± 3.2†	2.7 ± 2.4‡
With NP symptoms	13.6 ± 5	4.3 ± 3.2‡	2.7 ± 2.4‡
SDI, mean ± SD			
Without NP symptoms	1.4 ± 1.2	1 ± 1.1	1.2 ± 1.2
With NP symptoms	2.2 ± 1.4	1.2 ± 1.1‡	1.2 ± 1.2†
ACR 1982 criteria for SLE, no. (%)			
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)
Neurologic 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, no. (%)			
IgG aCL	8 (24.2)	5 (16.1)	2 (10)
IgM aCL	1 (3)	2 (6.5)	2 (10)
LAC	13 (39.4)	5 (16.1)	3 (15)
ANA	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)
Low C1q	3 (9.1)	1 (3.2)	1 (5)
Low C3	13 (39.4)	10 (32.3)	3 (15)
Low C4	12 (36.4)	6 (19.4)	5 (25)

* Three groups of systemic lupus erythematosus (SLE) patients were studied: those with neuropsychiatric SLE (NPSLE), those with NP symptoms unrelated to the SLE (non–SLE-related NP), and those with SLE without NP symptoms. A group of 36 healthy control subjects (32 women and 4 men; mean ± SD age 40.1 ± 11.8 years) was also studied. SLEDAI-2K = SLE Disease Activity Index 2000; SDI = Systemic Lupus International Collaborating Clinics/American College of Rheumatology (ACR) Damage Index; ANA = antinuclear antibody; aCL = anticardiolipin antibody; LAC = lupus anticoagulant; anti-dsDNA = anti–double-stranded DNA; ENA = extractable nuclear antigen.
† $P < 0.05$ versus NPSLE patients.
‡ $P < 0.001$ versus NPSLE patients.

values. The association between NPSLE syndromes and HPH values was assessed by independent t-test for every NPSLE syndrome present in >5 patients, taking into account a possible inequality of variances. A paired samples t-test was performed to test for significant mean differences in HPHs before and after treatment of active NPSLE. Statistical analysis was performed using SPSS version 20.0 software for Windows.

RESULTS

Patient selection and characterization. From all patients who were evaluated, 135 (73.8%) fulfilled the revised ACR criteria for SLE. In 59 of these patients (43.7%), a diagnosis of CNS NPSLE was established at the second visit. In the remaining patients, the NP symptoms were not directly attributable to SLE. After MRI evaluation, a total of 33 patients with CNS NPSLE and 31 with non–SLE-related NP met the criteria for our MTR study. The rest of the patients were excluded because of the presence of abnormalities on conventional MRI. Table 1 shows the clinical characteristics and autoantibody profiles of the study patients at the time of the first MRI. The values for the SLEDAI-2K with and

Table 2. White matter and gray matter MTR-HPHs measured in the study groups*

	No. of subjects	MTR-HPH, mean ± SD	
		White matter	Gray matter
Healthy controls	36	43.37 ± 5.11	10.01 ± 2.51
SLE	20	42.74 ± 6.22	10.02 ± 1.92
Non–SLE-related NP	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

* Peak height values were multiplied by 10,000 for readability. See Table 1 for definitions of the study groups. MTR-HPH = magnetization transfer ratio histogram peak height.

without NP symptoms and for the SDI with NP symptoms were significantly higher in the NPSLE group. No differences were found for the SDI values without NP symptoms. Among the patients diagnosed as having CNS NPSLE, 22 had inflammatory NPSLE and 11 had ischemic NPSLE. Fifty-four different ACR NP syndromes were established. Detailed clinical characteristics, including ACR NPSLE syndromes and NPSLE phenotype, are presented in Supplementary Table 1 (available on the *Arthritis & Rheumatology* web site at http://onlinelibrary.wiley.com/doi/10.1002/art.39653/abstract).

White and gray matter MTR-HPHs and NPSLE diagnoses. The mean ± SD values for the MTR-HPHs in the white and gray matter are shown in Table 2. The mean differences in the white and gray matter MTR-HPHs between the diagnostic groups are shown in Table 3. NPSLE patients with CNS involvement had significantly lower WM MTR-HPHs than did the healthy controls ($P < 0.001$) and the SLE patients ($P = 0.001$). No differences were found between the NPSLE group and the non–SLE-related NP group ($P = 0.114$). Patients with non–SLE-related NP had significantly lower WM MTR-HPHs than the healthy controls ($P < 0.001$). After adjustment according to the Tamhane procedure, no statistically significant differences between non–SLE-related NP patients and SLE patients were found ($P = 0.063$). Moreover, no statistically significant differences in WM values between the SLE patients and healthy controls were found. We did not find any significant differences in the mean GM MTR-HPHs between the various subgroups. Analyses controlling for differences attributable to disease duration, SLEDAI-2K, SDI, smoking status, hypertension, and aCL did not reveal any significant influence on the previous findings. Figure 1 shows the mean WM MTR histograms after correction for intracranial volume in the NPSLE, non–SLE-related NP, and SLE patients as well as in the healthy controls.

Table 3. Mean differences in white matter and gray matter MTR-HPHs between the study groups after the Tamhane procedure, by NPSLE diagnosis and NPSLE phenotype*

	White matter peak height		Gray matter peak height	
	Difference	P (95% CI)	Difference	P (95% CI)
NPSLE diagnostic groups				
NPSLE versus				
Healthy controls	−8.74	0.000 (−13.02, −4.47)†	−1.45	0.247 (−3.39, 0.48)
SLE patients	−8.12	0.001 (−13.38, −2.85)‡	−1.64	0.150 (−3.61, 0.32)
Non–SLE-related NP patients	−3.73	0.114 (−7.98, 0.51)	−1.24	0.654 (−3.63, 1.14)
Non–SLE-related NP versus				
Healthy controls	−5.01	0.000 (−8.24, −1.77)†	−0.21	1.000 (−2.35, 1.93)
SLE patients	−4.39	0.063 (−8.93, 0.16)	−0.39	0.997 (−2.56, 1.76)
SLE versus				
Healthy controls	−0.62	0.999 (−5.19, 3.94)	0.19	1.000 (−1.45, 1.84)
NPSLE phenotype groups				
Inflammatory NPSLE versus				
Healthy controls	−11.14	0.000 (−16.74, −5.54)†	−2.29	0.073 (−4.71, 0.12)
SLE patients	−10.52	0.000 (−16.93, −4.11)†	−2.48	0.044 (−4.93, −0.04)‡
Non–SLE-related NP patients	−6.13	0.023 (−11.71, −0.55)‡	−2.09	0.296 (−4.91, 0.72)
Ischemic NPSLE patients	−7.19	0.001 (−11.36, −3.02)‡	−2.53	0.276 (−5.96, 0.89)
Ischemic NPSLE versus				
Healthy controls	−3.94	0.165 (−8.75, 0.86)	0.24	1.000 (−2.89, 3.37)
SLE patients	−3.32	0.607 (−9.05, 2.41)	0.47	1.000 (−3.11, 3.19)
Non–SLE-related NP patients	1.06	0.999 (−3.73, 5.86)	0.44	1.000 (−2.92, 3.81)

* See Table 1 for definitions of the study groups. MTR-HPH = magnetization transfer ratio histogram peak height; 95% CI = 95% confidence interval.
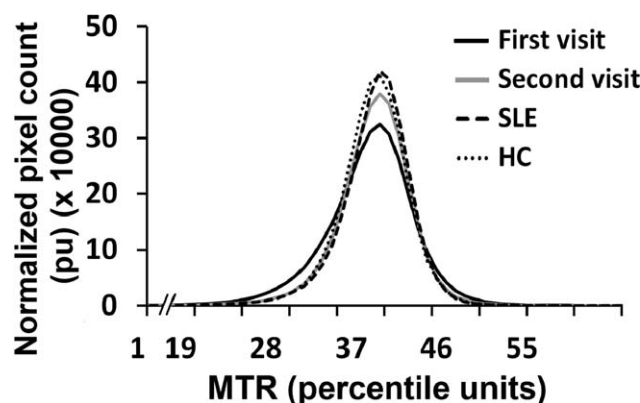† $P < 0.001$.
‡ $P < 0.05$.
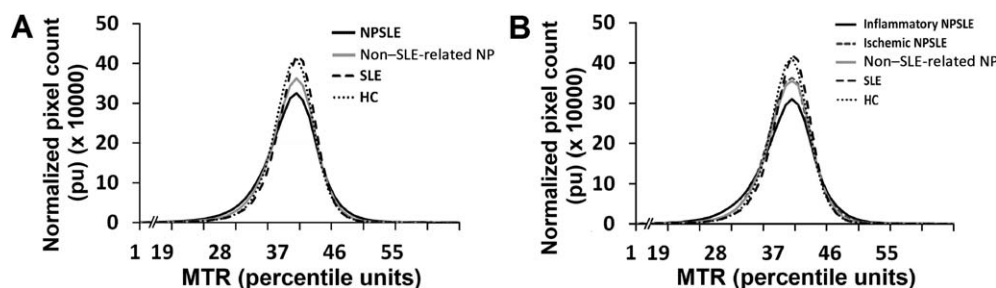
Figure 1. Average white matter magnetization transfer ratio (MTR) histograms. Mean MTR histograms after correction for intracranial volume are shown in **A,** patients with neuropsychiatric systemic lupus erythematosus (NPSLE), patients with NP symptoms unrelated to the underlying SLE (non–SLE-related 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 gray matter MTR peak heights and NPSLE phenotypes. Table 3 shows the mean differences in white and gray matter MTR-HTPs in the NPSLE phenotype groups. Patients with inflammatory NPSLE had significantly lower WM MTR-HPHs as compared with the healthy controls ($P < 0.001$), the SLE patients ($P < 0.001$), and the non–SLE-related NP patients ($P = 0.023$). Moreover, patients with inflammatory NPSLE had significantly lower WM MTR-HPHs as compared with patients with ischemic NPSLE ($P = 0.001$). No statistically significant differences were found for WM values when we compared ischemic NPSLE patients with healthy controls, non–SLE-related NP patients, or SLE patients. Patients with inflammatory NPSLE also had significantly lower GM MTR-HPHs as compared with the SLE patients ($P = 0.044$), but we found no other differences as compared with the other subgroups. We did not find any statistically significant differences for the GM values when ischemic NPSLE patients were compared with the healthy controls, the non–SLE-related NP patients, and the SLE patients. Analyses controlling for differences attributable to disease duration, SLEDAI-2K, SDI, smoking status, hypertension, and aCL did not reveal any significant influence on the previous findings. WM MTR histograms in the 5 study groups are shown in Figure 1B.

White and gray matter MTR peak heights and NPSLE syndromes. Independent *t*-test analyses were performed for every NPSLE 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 lower WM ($P = 0.033$) and GM ($P = 0.029$) MTR-HPHs. We also found an association between lower WM MTR-HPHs and cognitive disorder ($P = 0.047$) as well as mood disorder ($P = 0.025$). We did not find any association between the GM MTR-HPHs and either

cognitive disorder or mood disorder. Cerebrovascular disease was also associated with higher WM MTR-HPHs ($P = 0.006$). We found no associations between MTR-HPHs and headache or seizure.

White matter MTR peak heights and clinical changes. Of the 20 NPSLE patients considered to have active CNS disease during the first visit, 11 were classified as improved after treatment, 7 as stable, and 2 as worse. The mean ± SD WM MTR-HPHs in all patients at the first visit was 31.51 ± 7.83, and at the follow-up visit, this value had increased to 39.07 ± 6.56. Figure 2 shows WM MTR histograms before and after treatment (after correction for intracranial volume). In all NPSLE patients with clinical improvement, the mean WM MTR-HPH increased by 9.81 ± 5.94 (range 5.81 to 13.81) ($P < 0.000$). The mean ± SD difference in WM MTR-HPHs in patients classified as stable at the second visit was 2.48 ± 4.65 (range 1.81–6.79) ($P = 0.207$). In the

Figure 2. Average white matter magnetization transfer ratio (MTR) histograms after correction for intracranial volume are shown in patients with active neuropsychiatric systemic lupus erythematosus (NPSLE) at baseline (first visit) and in the same NPSLE patients after treatment (second visit). Baseline MTR histograms for SLE patients without NP symptoms and for healthy control (HC) subjects are included for comparison. pu = percentage units.

2 patients whose condition worsened, a decrease in the WM MTR-HPH between the first and second MRI was observed: -10.32 ± 0.41 (range -14.01 to -6.63) ($P = 0.018$).

DISCUSSION

This study is the first to show that NPSLE patients with an inflammatory phenotype have significantly lower WM MTR-HPHs than do ischemic NPSLE, 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 NPSLE.

Inflammatory NPSLE 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 NPSLE 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 NPSLE, 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 NPSLE, mainly seen in those with cerebrovascular symptoms, were lower than those in the healthy controls and significantly higher than those in patients with inflammatory NPSLE 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 NPSLE patients, probably reflecting residual effects or WM-specific and irreversible changes in patients with past inflammatory NPSLE.

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 NPSLE 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 NPSLE 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 between SLE patients and healthy controls, which may suggest that each technique identifies different aspects of the microstructural changes in the brains of SLE and NPSLE patients. As previously reported, no differences between NPSLE patients and non–SLE-related NP patients were found, probably because the NPSLE group included both ischemic and inflammatory NPSLE 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 NPSLE 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 NPSLE. As mentioned above, nonspecific microstructural changes of the brain tissue as measured 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 NPSLE 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 NPSLE as compared with those with SLE and a trend as compared with healthy controls. The difference between NPSLE 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 NPSLE 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 NPSLE (8,9). Due to partial volume effects, the voxels analyzed in the parenchymal 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 prevalence and the high heterogeneity of NPSLE. We therefore cannot draw definite conclusions concerning the relationship between the MTR-HPH findings and NPSLE syndromes. Furthermore, due to matters of referral, some of the patients with inflammatory NPSLE were evaluated in the NPSLE clinic once they had started the immunosuppressive therapy. This may explain the higher variance in the NPSLE group, and we believe that inflammatory NPSLE 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 NPSLE 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 NPSLE 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 NPSLE 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 NPSLE. This study also confirmed the usefulness of this technique in the detection of cerebral changes in NPSLE 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. Further 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 NPSLE and with the monitoring of future treatment trials.

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AUTHOR CONTRIBUTIONS

All authors were involved in drafting the article or revising it critically for important intellectual content, and all authors approved the final version to be published. Dr. Magro-Checa had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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