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Should frequent MRI monitoring be performed in natalizumab-treated MS patients? A contribution to a recent debate

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Abstract

Background: Brain magnetic resonance imaging (MRI) is the most effective surveillance tool for the detection of asymptomatic progressive multifocal leukoencephalopathy (PML). However, the optimal frequency for routine MRI surveillance is under-investigated.

Objective: To understand whether, upon their first MRI appearance, PML lesions present a difference in volume when comparing patients who frequently underwent MRI surveillance (3/4 months) with those who were assessed at longer intervals (6/12 months) and to understand the impact of the volume of lesions on clinical outcome.

Methods: The data of patients included in the Italian PML cohort were retrospectively analysed. Patients who had all the pre-diagnostic MRI scans available ($n = 37$) were included. The volume of PML lesion was calculated by manually outlining the PML lesion.

Results: Compared with patients who underwent MRI examination at least every 4 months, patients who were assessed less frequently had a lesion of significantly higher volume (median: 2567 (883–3583) vs. 664 mm^3 (392–963) $p = 0.006$) and suffered a higher rate of disability (median: 2.25 expanded disability status scale points (–2.5 to 8) vs. 0.5 (–1 to 2.5) $p = 0.004$).

Conclusion: The positive clinical outcome of patients undergoing frequent MRI surveillance and the small volume of the PML lesion upon first appearance justify a frequent surveillance using MRI in patients at high risk of PML.

Keywords: Natalizumab, progressive multifocal leukoencephalopathy, lesion volume, high frequency MRI

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Introduction

Progressive multifocal leukoencephalopathy (PML) is an opportunistic infection of the brain occurring in immune-compromised patients. In immune-competent patients, as in multiple sclerosis (MS), the risk of PML is potentially high (incidence ranging from 1–100 to 1–1000 depending on the presence of risk factors). This risk increases when patients are treated with natalizumab (NTZ),[1] a monoclonal antibody that binds to α4β1 integrin and prevents lymphocytes from crossing the blood–brain barrier.[2] For a diagnosis of definite PML, the diagnostic criteria suggested by the American Academy of Neurology[3] require the presence of JCv (the virus causative of PML) in the cerebrospinal fluid (CSF). In addition, brain magnetic resonance imaging (MRI) findings and clinical symptoms suggestive of PML are needed as well. This diagnostic pathway, however, has recently been challenged, as evidence is emerging that patients with smaller lesion volumes are more likely to be asymptomatic, with undetectable JCv DNA copies in CSF.[4–7] In this scenario, MRI is crucial for the early diagnosis of PML.

In the absence of an effective treatment for PML, the timely withdrawal of NTZ is essential to restore the immune response and to reduce the spreading of infection.[2] In fact, smaller lesion volume and/or

asymptomatic presentation leads to improved survival and functional outcome.[5,8,9] As MRI is, to date, the most effective surveillance method for the detection of asymptomatic PML,[10] it has been introduced as a screening tool.[11] The PML screening protocol that was initially proposed, which includes fluid attenuated inversion recovery (FLAIR), T2-weighted and T1-weighted images,[12] has been incorporated into expert guidelines.[13,14] Given its relevance, this protocol has also been included in the updated recommendations from the European Medical Agency (EMA) to assist the early identification of PML with NTZ.

Nevertheless, the best frequency for routine MRI surveillance is still an underinvestigated topic. A debate in the literature has consequently emerged. On one hand, some authors have suggested that frequent MRI examinations might offer the opportunity to identify emerging PML prior to the onset of clinical symptoms.[15,16] On the other hand, there are some concerns that overreliance on MRI might jeopardise clinical vigilance, thus increasing the diagnostic uncertainty, which could lead, in some cases, to unnecessary cessation of therapy.[17]

Recent expert guidelines have not helped to fully disentangle this diatribe. Although it is recommended by all the existing guidelines that patients at increased risk of PML should be monitored at shorter intervals, the optimal frequency of MRI surveillance is still under discussion. For instance, The MAGNIMS guidelines[14] and expert opinions recently published by Major et al.,[2] recommend high-frequency brain MRI (every 3 or 4 months) in patients at higher risk of developing PML. Similarly, in the article by McGuigan et al.,[13] an MRI scan every 6 months is the minimum suggested for patients with index < 1.5, while a 3–4-month interval is suggested for an index > 1.5. Furthermore, Montalban et al.[18] and Traboulsee et al.[19] recommend to perform MRI on a 3–6 monthly basis in high-risk patients (i.e. JCv antibodies positivity and therapy duration > 18 months). Finally, the EMA recommends an MRI scan every 3 to 6 months in patients at higher risk (e.g. JCv index > 1.5 and therapy duration > 2 years). The recommendations from these guidelines are based on few clinical evidences suggesting a strict MRI pharmacovigilance.[8,10,11,20–23]

To date, no studies have defined the best MRI frequency for PML surveillance in MS. This is also clearly stated in a commentary on the best MRI scanning interval.[15] This article underlined how the suggested scanning frequency is currently based on class IV evidence, and noted that unnecessarily frequent MRI scans would entail remarkable costs for any national health system. It is, therefore, of the utmost importance that additional data are provided to support or contrast these recommendations.

This study aims to retrospectively analyse the Italian PML data set[9,24] to investigate whether MRI frequency has an impact on both lesion size upon first detection of PML by MRI (measured by both lesion dissemination and lesion volume) and on the clinical outcome.

Materials and methods

Patients and data collection

The data of patients included in the Italian PML cohort[9,24] were retrospectively analysed. This cohort encompassed all the Italian patients who were diagnosed with NTZ-related PML up to May 2018. Patients were included in the current study if they met either the AAN diagnostic criteria for PML[3] or the recently proposed diagnostic criteria for PML[6] and if their MS/PML history was fully available.

The following data are considered in the present study: gender; age at MS onset; seropositive status (JCv index of 14 patients, as the test was not available before 2013); age at PML insurgence; expanded disability status scale (EDSS)[25] before NTZ first administration, at PML diagnosis and at 12-months follow-up; prior immune-suppressant use; number of NTZ infusions; CSF JC viral copies/ml at PML diagnosis; clinical symptoms at PML diagnosis; frequency of surveillance by MRI during NTZ treatment (every 12, 8, 6, 4 or 3 months).

The retrospective analysis of patients' data was approved by the Ethical Committee of the Spedali Civili of Brescia and was conducted in accordance with the ethical standards laid down in the Declaration of Helsinki (1964) and its later amendments.

Localisation of the lesion

All collected images were acquired using a 1.5T scanner. Although brain MRI scans were based on local protocols, T1 weighted, T2 weighted, FLAIR sequences were acquired for each of the 37 patients. Furthermore, diffusion weighted images (DWI) were acquired in 29 (78.37%) and post contrast T1-weighted images in 34 (91.89%) patients. Lesions at their first MRI appearance were identified in consensus by two expert neuroradiologists (MC and SG) according to shared neuroradiological criteria;[3,26–28] the lesion dissemination was rated as unilobar, multilobar or

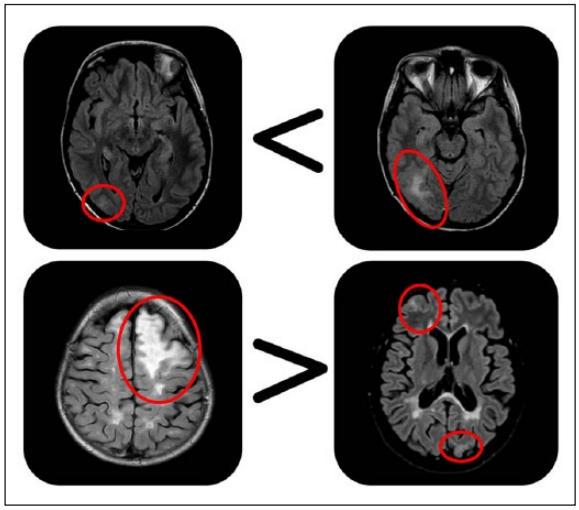

Figure 1. The importance of lesion's volume calculation. Upper panel: longitudinal lesion of the same patient: the lesion remain unilobar over time, but its volume might clearly increase. Lower panel: comparing the PML lesions of two patients: the lesion's volume at PML diagnosis is larger in patients with unilobar lesion than in patients with a widespread lesion.

widespread,[29] consistent with previous literature on PML.[4,5] In order to verify PML lesion identification, previous and subsequent MRI scans were also reviewed by the raters.[5]

Calculation of lesion volume

We believe that the lesion dissemination is not an accurate proxy of the lesion's real volume. Indeed, we observed that (1) the lesion might remain unilobar over time, but its volume might clearly increase (for exemplificative purpose, see Figure 1, upper panel); (2) the lesion's volume might be bigger in a unilobar lesion than in a widespread lesion (for exemplificative purpose, see Figure 1, lower panel). Thus, we calculated the lesion's volume. In particular, for each patient the lesion extent and location were assessed using the MRI scan where the PML appeared for the first time. In accordance with previous relevant literature,[5] the detection of the lesion was performed on axial FLAIR images (94.59% of cases) or when axial FLAIR was unavailable, on sagittal FLAIR (5.40% of cases). This is because FLAIR has the highest sensitivity for the detection of PML lesions.[29] In order to make the lesion volume directly comparable between patients, two raters manually outlined in consensus the PML lesions on the axial images of the Montreal Neurological Institute (MNI) template using the MRIcron software[30,31] (available at http://www.mricro.com/mricron). The manual outline of the lesion was then verified by a third rater. This technique is widely used in brain lesion imaging research.[4,32,33] Superimposing each patient's lesion onto the standard brain allowed us to estimate the total brain lesion volume (in cubic centimetres) removing inter-subject variability in total head size and total intracranial volume. Importantly, this strategy also allowed us to mitigate the effect of different acquisition parameters (i.e. slice thickness) in each site on the lesion volume.

Statistical analysis

Results are reported as mean with standard deviation (SD) or median with interquartile range (IQR) for quantitative characteristics. Quantitative characteristics were compared between patients who underwent MRI surveillance frequently (every 3 or 4 months) or infrequently (from 6 to 12 months), using independent samples Student's t test or Mann–Whitney test, depending on whether the data were or were not normally distributed. In particular, MRIs performed every 3–4 months were considered frequent[2,14] following previous observation in the same cohort that the asymptomatic pre-diagnostic PML phase is about 5 months long[4] (up to 41 weeks according to previous literature).[2] For explorative purposes, analyses were also repeated dividing the patients into four groups: patients who underwent MRI every 3 months (group 1, $n=5$), every 4 months (group 2, $n=11$), every 6 months (group 3, $n=10$) and >6 months (group 4, $n=11$).

The standardised mean difference between groups of MRI frequency was calculated according to the Yang and Dalton's[34] method for all characteristics. To adjust the lesion's volume comparison between frequent and infrequent MRI groups, the baseline characteristics that showed a quote of unbalance >0.10 in standardised mean difference were considered.[35] These characteristics were selected to enter in a stepwise linear regression model and those with $p<0.15$ were included in the final multivariable model. For clinical reasons, the previous use of immune suppressant and the presence of brainstem lesions were forced to enter into the model. Brainstem lesions were included in the model as the lesion's volume within the brainstem is constrained by the small volume of the structure and the lesion cannot expand beyond anatomical boundaries. The lesion's volume was log-transformed and used as dependent variable.

The association of MRI frequency with overall survival was tested using the chi-square test, while the association with delta EDSS was tested using the of non-parametric Mann–Whitney test. As for the

lesion's volume, a multivariable linear regression was used to adjust delta EDSS comparison between frequent and infrequent MRI groups using the same approach previously described. EDSS change between the beginning of NTZ administration and 12-month follow-up was transformed in ranks before applying a linear model. Finally, in order to provide a preliminary explanation for the EDSS results between groups, the localization of the lesion was described. In particular, the number of patients with a unilobar lesion in the non-motor regions of the frontal lobe has been recorded, as these lesions are associated with symptoms that are less clinically evident.[36]

Stata (v.14; StataCorp.) was used for the computation.

Results

Clinical features

Thirty-seven patients met the inclusion criteria. Twenty-seven patients (72.97%) met the AAN diagnostic criteria for definite, seven patients (18.91%) for probable (as they were asymptomatic at PML diagnosis) and one patient (2.70%) for possible PML. This last patient has undetectable JCv in the CSF, while MRI lesions (in the frontal motor regions) and clinical symptoms (impairment in the movement of right hand) suggestive of PML were present. Two patients (5.40%) did not meet the AAN criteria for PML as they were asymptomatic at PML diagnosis and the JCv was not detectable in the CSF. However, these patients were included in the current study in accordance with the most recently published data:[6,26,29] no disease activity during the treatment with NTZ was documented and the clinical and radiological longitudinal evolution confirms the one expected for PML in patients at high risk of developing PML. In particular, the lesion's volume increased over time, contrast enhancement suggestive of immune reconstitution inflammatory syndrome (IRIS) occurred from 2 to 3 months after NTZ withdrawal[24] and symptoms compatible with PML[36] emerged during the clinical course of the disease. These patients had been already included in our cohort in a previous publication.[4]

For the subjects with a JCv index ($n=14$), the mean JCv index was 3.08 ± 0.94 (median: 3.1 (1.52–5.27)). Sixteen patients underwent MRI frequently, while 21 were scanned by MRI at longer intervals. The demographic and clinical features of patients are presented in Table 1. Although not significant ($p=0.14$), the percentage of asymptomatic patients at PML diagnosis is clinically higher in the group that received frequent scans (35.3%) as compared to those who received infrequent (13.6%) scans. Furthermore, the number of JCV DNA viral copies/ml at PML diagnosis is lower in the group of patients who underwent frequent (37 (10–117)) as compared to infrequent (355 (50–1307)) MRI ($p=0.002$) scans.

Lesion dissemination

The MRI sequences available and used in this study in both groups are summarised in Table 2. At the first MRI appearance, PML lesions were unilobar in 27 out of 37 (72.97%) participants. The lesions were significantly more likely to be unilobar in the group of patients who underwent frequent (15/16, 93.75%) rather than infrequent (12/21, 57.14%, chi-square = 6.17, $p=0.013$) MRI scans. When MRI frequency was considered in four categories, multilobar lesion at first PML appearance were detected in one patient out of 11 in the 4-month frequency group (9.09%), 6/10 of the 6-month frequency (60%) and 4/11 of the >6-months frequency (36.4%) group.

Lesion volume

Patients with frequent MRI had a significantly smaller volume of lesion at PML first MRI appearance ($p=0.008$). All the clinical and the demographic characteristics considered (reported in Table 1) showed $p>0.20$ in the linear regression model. The smaller PML lesion volume among patients with frequent MRI was confirmed ($p=0.006$) after adjustment for previous use of immune suppressants ($p=0.59$) and brainstem lesions ($p=0.32$; Figure 2). When MRI frequency was considered in four categories, a significantly higher volume was observed for >6 months (mean: 11496 mm^3) versus both 3 months (mean: 599.3 mm^3, $p=0.005$) and 4 months (mean: 1057 mm^3, $p=0.012$) without significant differences between 3 and 4 months ($p=0.53$). A trend was observed versus a higher volume for 6 months frequency (mean: 3501 mm^3) when compared with 3 months ($p=0.076$) and 4 months ($p=0.17$) frequency. A similar result was observed comparing lesion volume of 6 months and >6 months ($p=0.17$). Results are presented in Figure 3. These latter results must be considered with caution given the very small number of subjects within each group.

Clinical outcome

Among 21 patients with infrequent MRI, three (14.2%) had a fatal event, while no fatal events were observed in the frequent MRI group. EDSS change from the initial NTZ administration to 12 months follow-up, adjusted for previous use of immune-suppressant drugs ($p=0.095$) and ARR pre-treatment ($p=0.023$),

Table 1. Demographic and clinical variables in patients who underwent MRI frequently or infrequently.

	Frequent MRI ($n=16$)	Infrequent MRI ($n=21$)	Standardised difference	p
Age at MS onset[a]	25.2 (7.3)	27.5 (9.1)	0.29	0.38
Age at PML diagnosis[a]	39.7 (7.8)	39.8 (9.7)	0.008	0.98
Gender (female)[b]	12 (70.6%)	13 (59.1%)	0.24	0.52
ARR before NTZ[a]	2.67 (0.78)	1.77 (0.87)	1.08	**0.011**
ARR during NTZ[a]	0.36 (0.48)	0.06 (0.22)	0.81	0.026
Disease duration at PML (years)[a]	14.5 (9.4)	12.2 (5.6)	0.23	0.48
Viral load in the CSF (copies/millilitre)[c]	37 (10–117)	355 (50–1307)	1.17	**0.002**
Number of NTZ infusions[c]	43.4 (21.6)	43.2 (19.2)	0.006	0.98
Number of asymptomatic patients[b]	6 (35.3%)	3 (13.6%)	0.52	0.14
EDSS at NTZ beginning[c]	4 (2–6)	3 (2–4)	0.35	0.25
EDSS at PML diagnosis	4 (3–6)	4 (2.5–6)	0.12	0.68
Previous use of immune suppressant (yes)[b]	4 (23.5%)	7 (31.8%)	0.19	0.73
Presence of contrast enhancing lesions (yes)[a]	1 out of 14 (7.14%)	5 out of 20 (25%)	0.50	0.36
Lesion volume (mm^3)[c]	644 (392–963)	2567 (883–3583)	0.99	**0.008**
MRI surveillance frequency	$n=11$ every 4 months $n=5$ every 3 months mean 3.7 ± 0.5	$n=7$ every 12 months $n=4$ every 8 months $n=10$ every 6 months mean 8.4 ± 2.7		

MS: multiple sclerosis; ARR: annualised relapse rate; NTZ: natalizumab; EDSS: expanded disability status score; CSF: cerebrospinal fluid; PML: progressive multifocal leukoencephalopathy; MRI: magnetic resonance imaging.
[a]number denotes average (standard deviation); statistical significance was evaluated using two independent sample *t* test.
[b]number denotes row number (percentage); statistical significance was evaluated using chi-square.
[c]number denotes median (interquartile range); statistical significance was evaluated using Mann–Whitney *U* test.
Bold denotes statistical significance.

was significantly lower ($p=0.004$) in patients with frequent MRI (mean change: 0.91 (SD: 1.03); median: 0.5 (range: –1 to 2.5)) than in patients with infrequent MRI scans (mean change: 2.7 (SD: 2.7); median: 2.25 (range: –2.5 to 8); Figure 4). When MRI frequency was considered in four categories, a fatal event was observed in the > 6 months MRI frequency group and two fatal events in the 6 months MRI frequency group. Furthermore, a significantly higher EDSS change was observed for 6 months (mean: 3.5, median: 3.5) versus both 3 months (mean: 0.7, median: 0.5; $p=0.012$) and 4 months (mean: 1.05, median: 1; $p=0.01$) without significant differences between 3 and 4 months ($p=0.85$). A trend towards a higher EDSS change for > 6 months frequency (mean: 2, median: 1.5) was observed when compared with 3 months ($p=0.12$) and 4 months ($p=0.13$) frequency. No significant differences were observed when comparing 6 versus > 6 months ($p=0.23$). The results are represented in Figure 5. The latter results must be considered with caution given the very small number of subjects within each group.

Lesion localisation

The number of unilobar frontal lesions did not differ between patients who underwent MRI frequently (6/16, 37.5%) or infrequently (7/21, 33.3%, chi-square = 0.611, $p=0.25$). When MRI frequency was considered in four categories, unilobar frontal lesions were present in 3/5 (60%) patients within the 3-months group; 3/11 (27.27%) within the 4-months; 2/10 (20%) within the 6 months and 5/11 (45.45%) within the > 6-months group.

Discussion

Despite the critical relevance of MRI in the early diagnosis of PML, the optimal frequency of MRI surveillance is still undefined. Experts guidelines[2,13,14,18,19] are trying to overcome this gap in the literature. Their recommendations are based on both the opinions of experts and on extensive reanalysis of the available literature that provides mostly indirect support to the clinical usefulness of frequent MRI. So far, few retrospective studies have indirectly investigated the

Table 2. Details on MRI sequences available and used in this study.

MRI sequence	Frequent MRI ($n=16$)			Infrequent MRI ($n=21$)		
	Axial	Sagittal	Coronal	Axial	Sagittal	Coronal
Available MRI sequences						
FLAIR	15(93.75%)	12(75%)	5(31.35%)	20(95.23%)	11(53.38%)	9(42.85%)
T2	12 (75%)	1(6.25%)	7(43.75%)	16(76.19%)	11(53.38%)	2(9.52%)
T1	13(81.25%)	6(37.5%)	2(12.5%)	16(76.19%)	8(38.09%)	3(14.28%)
DWI	11(68.75%)	0	0	18(85.71%)	0	0
T1 with contrast	14(87.5%)	6(37.5%)	8(50%)	20(95.23%)	12(57.14%)	13(61.90%)
MRI sequence used for PML lesion volume measurement						
Axial FLAIR	15 (93.75%)			20 (95.23%)		
Sagittal FLAIR	1 (6.25%)			1 (4.77%)		

Numbers denote the row number of patients (percentage).
MRI: magnetic resonance imaging; PML: progressive multifocal leukoencephalopathy; FLAIR: fluid attenuation inversion recovery; T2: T2-weighted images; T1: T1-weighted images; DWI: diffusion weighted images.

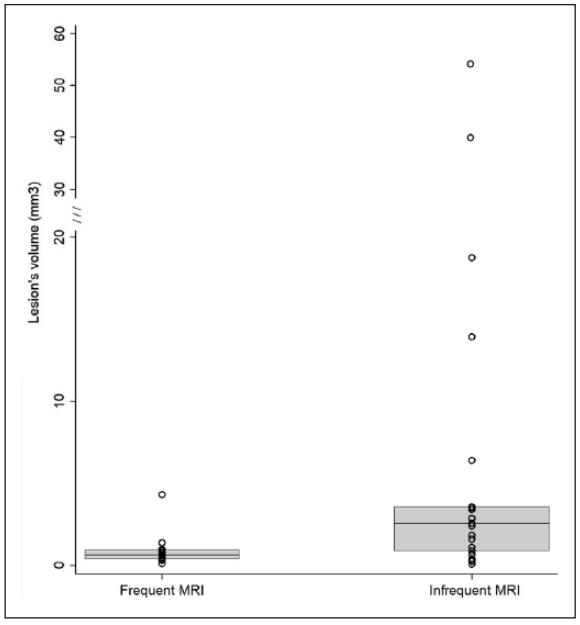

Figure 2. PML lesion volume at the first appearance.
Boxplot that represents the extent and overlap of brain lesions in patients who underwent MRI frequently and infrequently ($p=0.006$). The black line is the median, and the box represents the 25th and 75th percentile.

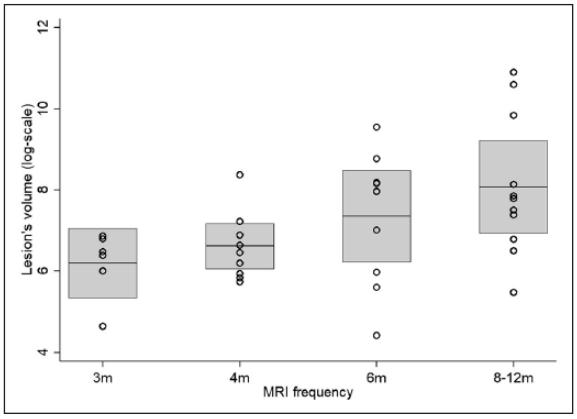

Figure 3. PML lesion volume at the first appearance in four groups.
Boxplot that represents the extent and overlap of brain lesions in patients who underwent MRI every 3, 4, 6 and > 6 months. The black line is the median, and the box represents the 25th and 75th percentile.

impact of MRI frequency.[8,27,28] Case reports suggesting a role of frequent MRI surveillance in good clinical outcomes are also available.[10,11,20–22] However, these data need further support. The present study enriches the current literature providing retrospective evidence to support the introduction of MRI surveillance performed at least every 4 months to identify localised (i.e. unilobar) and smaller volume PML lesions. More importantly, the study also demonstrates the influence of strict MRI surveillance on longitudinal disability outcome.

Despite CSF markers, such as the CSF index of JC virus, have been proposed for the early diagnosis of PML,[37] a frequent MRI surveillance is undoubtedly useful for the detection of PML lesions in the asymptomatic stage, promoting timely NTZ withdrawal and avoiding a wide diffusion of the virus in the brain. Moreover, patients undergoing MRIs at least every 4 months are more likely to be asymptomatic with a lower, sometimes undetectable, viral load. This would, in turn, explain previous studies suggesting that asymptomatic patients have a better outcome.[8,9]

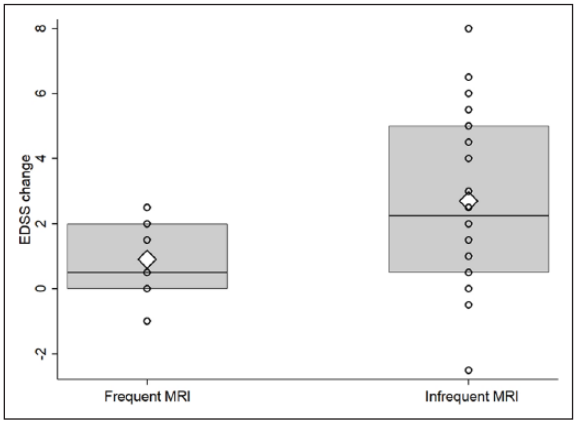

Figure 4. Disability change from NTZ beginning to 12-months follow-up.
Boxplot that represents the EDSS change at 12 months in patients who underwent MRI frequently and infrequently ($p = 0.004$). The black line is the median, and the box represents the 25th and 75th percentile, while the white diamond represents the mean.

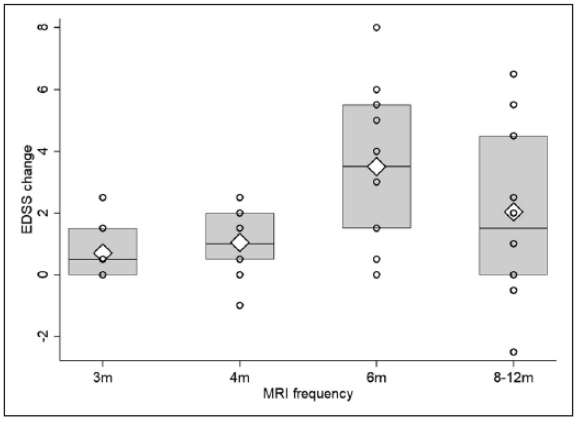

Figure 5. Disability change from NTZ beginning to 12-months follow-up in four groups.
Boxplot that represents the EDSS change at 12 months in patients who underwent MRI every 3, 4, 6 and > 6 months (m = months). The black line is the median, and the box represent the 25th and 75th percentile, while the white diamond represent the mean.

There is still no agreement between experts on the optimal frequency for MRI surveillance[2,13,14,18,19] and the frequency varies in different countries. For instance, in U.S. best practice recommendations, published in 2009[38] by an expert panel, recommended MRI examination at least annually in NTZ-treated patients. In fact, several panel members reported that they performed a routine MRI 6 months after the beginning of treatment and then on an annual basis. However, more frequent MRI surveillance has been suggested in Europe.[39] Our data showed a higher percentage of asymptomatic patients in those who underwent more frequent MRI. This gives rise to the hypothesis that the frequency of MRI surveillance could account for geographical differences in the distribution of asymptomatic PML patients, as a higher percentage of asymptomatic patients has been reported in Europe (86.7%) than in the Unites States (13.3%).[8] Although the 2009 U.S. recommendations were updated in 2016[19] to suggest a more frequent MRI monitoring for high-risk patients, the older guidelines will have influenced the percentage of asymptomatic patients reported in the last 10 years.

This study has also underlined a clear positive prognostic impact of at least every 4-month MRI surveillance. Compared with the group of patients who underwent infrequent MRI, no fatal outcome was reported with frequent MRI. These patients also suffer lower disability accumulation over a 1-year follow-up, which is both statistically and clinically relevant. A previous study from the Italian PML working group found a strong correlation between viral copies at PML diagnosis and the disability change during the PML course.[9] As we found higher viral load at PML diagnosis in patients who underwent MRI at longer intervals, it is not surprising that these patients are also the ones with the worst clinical outcome (and a larger lesion volume). Also, three deaths occurred only in the group of patients with infrequent MRI. It is also likely that the frequency of MRI surveillance could also account for the different geographical distribution of deaths of PML patients:[40] historically, a higher mortality rate has been reported in United States (41%), where former guidelines recommended to perform an MRI scan at least annually,[38] while the rate is much lower in Europe (15%), where, according with guidelines, MRI scan is performed more frequently.[39]

It is also worth noting that, when MRI frequency was considered in four categories, patients with MRI frequency > 6 months seem to have *prima facie* a better clinical outcome compared with patients with MRI frequency of 6 months (even if not statistically significant). This result has two complementary explanations. First, patients in the > 6-months group have a higher prevalence of frontal unilobar lesion. It is well known that lesion within the non-motor regions of the frontal lobe are associated with symptoms that are less clinically evident[36] and might go unnoticed without a formal neuropsychological assessment.[36] For this reason, their symptomatology has a small impact on the EDSS total score. Second, from the current data, it is not possible to estimate the exact timing of PML insurgence. It is, thus, possible to speculate that the PML of patients performing MRI every > 6 months that presented with a small lesion volume and

exhibited only small worsening of clinical symptoms emerged only few months before the surveillance MRI, by chance. Although not statistically significant due to the small number of patients included, the EDSS results are still clinically meaningful. Indeed, patients undergoing MRI every 3 or 4 months have a mean EDSS worsening of 0.7 and 1 point, respectively, while patients undergoing MRI every > 6 months have a mean EDSS worsening of 2 points.

Although a comprehensive clinical assessment, both neurological and neuropsychological,[36] to identify the most insidious onset, remains pivotal to identify PML at the earliest symptomatic stage, frequent MRI monitoring offers the potential to identify asymptomatic emerging PML.[16] This is even more relevant as there is now convincing evidence to suggest that the earlier PML is detected, the smaller the lesion volume. As lesion volume is the only predictive marker with respect to JCv detection in the CSF,[5] patients with very small lesions may be asymptomatic with an undetectable level of virus in the CSF,[4,5] hampering the classical diagnosis of PML based on the AAN diagnostic criteria.[3] It is thus increasingly clear that the presence of a new lesion at MRI scan can be the only red flag for PML and this condition could last for 5 months on average,[4] and ranging up to 41 weeks.[2] Seven patients (five patients in the Dutch-Belgian[6] cohort and two in the Italian cohort)[4] have been already diagnosed with PML on the basis of radiological data only, supporting the pivotal role of MRI surveillance in the early detection of PML. The latency of PML clinical manifestation[4] supports the usefulness of MRI surveillance at least every 4 months in JCV positive patients, in order to provide a more strict pharmacovigilance, while continuing treatment with NTZ. Neuroradiologists should be aware of the relevance of very small PML lesions and how to differentiate PML from MS lesions[26] in asymptomatic patients. Summarising, a frequent MRI surveillance plan for patients with positive JCv would potentially lead to the early diagnosis of PML, which would strongly influence the clinical course of the adverse event. Being able to early diagnose PML through MRI, clinicians could be able to withdraw NTZ when the PML lesion is small, the viral load is low and the patient is clinically asymptomatic. This would affect the mortality rate and long-term residual disability. The current results support the emerging idea that revised PML diagnostic criteria are needed.[6]

Limitations

Besides the small number of PML patients included and the retrospective nature of this study, a limitation is that the impact of the JCv index has not been analysed. Unfortunately, this information was not available before 2013 for most patients ($n = 23$ out of 37). Furthermore, the available guidelines[2,13,14,18,19] adjust the suggested frequency of MRI surveillance to the PML risks factors (as previous immune suppressant use, NTZ therapy length and JCv index). The absence of a JCv index in our database prevents us from adjusting the results to the PML risk factors. Furthermore, as this retrospective study included patients from different Italian sites, MRI acquisition protocols are not standardised, as in previous research.[4,5] However, we are confident that the identification of the lesions by consensus has mitigated the bias introduced by the different scanning protocols. Furthermore, the lesion manual tracing method that we applied has been specifically selected to mitigate the effect of different acquisition parameters on the lesion volume.

Conclusion

The results provide the first evidence to justify the usefulness of MRI surveillance every 3 to 4 months for patients at risk of PML. Indeed, the current results clearly enrich the available literature as they suggest that, compared with patients who underwent MRI surveillance infrequently, the patients who were frequently examined present (1) narrower lesion dissemination and a smaller volume of lesions of PML at first MRI appearance; (2) a higher likelihood of being asymptomatic and a lower viral load at PML diagnosis; (3) lower mortality rate and (4) lower worsening of the clinical condition at the last follow-up. This knowledge might be implemented in future guidelines to monitor NTZ-treated patients.

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