APPLICATIONS OF MAGNETIC RESONANCE IMAGING - BASED BRAIN VOLUMETRY IN THE DIAGNOSIS AND FOLLOW - UP OF PATIENTS WITH MULTIPLE SCLEROSIS

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Abstract

Multiple sclerosis (MS) is a chronic demyelinating disease of the central nervous system (CNS) characterized by a widely variable clinical manifestation and course. Although MS is generally considered a disease of the white matter (WM), pathology is also found in the gray matter (GM). Axonal loss and neurodegeneration occur early and can lead to permanent neurological and cognitive impairment.

Magnetic Resonance Imaging (MRI) offers excellent anatomical - structural information and since 2001, MRI has been incorporated in the diagnostic workup of patients with a clinical suspicion of MS (1). Changes in brain volume, detected from the early stages of MS and proceeding throughout the course of disease, may offer an accurate measure of neurodegeneration and tissue damage. Qualitative (i.e., visual) interpretation of structural brain images can detect only macroscopic changes and shows a low level of interobserver agreement. On the other hand, advanced MRI techniques –such as MRI-based brain volumetry– developed in the recent years through the advances in computational technology have greater sensitivity than conventional MRI and allow quantitative assessment of structural brain images. Since manual identification and measurement of MS lesions on MRI can be extremely time-consuming and subjective, multiple semi-automated and automated methods for the determination of overall and regional brain volumes and of "lesion load" have been suggested. Nonetheless, although these methods seem sensitive and reproducible, their role should be mainly supportive to that of visual assessment of brain structural images, since variable factors (e.g. pseudoatrophy) can have a confounding influence on measurements.

After a brief overview of the different volumetric MRI techniques, this review will assess the clinical use of MRI-derived brain volumetry in the diagnosis, follow-up and monitoring of treatment effects in patients with MS.

Key words: magnetic resonance imaging, volumetry, Radiologically Isolated Syndrome, Clinically Isolated Syndrome, multiple sclerosis

ΚΛΙΝΙΚΗ ΕΦΑΡΜΟΓΗ ΤΗΣ ΟΓΚΟΜΕΤΡΗΣΗΣ ΤΟΥ ΕΓΚΕΦΑΛΟΥ ΜΕ ΕΙΔΙΚΕΣ ΜΕΘΟΔΟΥΣ ΑΠΕΙΚΟΝΙΣΗΣ ΜΑΓΝΗΤΙΚΟΥ ΣΥΝΤΟΝΙΣΜΟΥ ΣΤΗ ΔΙΑΓΝΩΣΗ ΚΑΙ ΠΑΡΑΚΑΚΟΛΟΥΘΗΣΗ ΤΩΝ ΑΣΘΕΝΩΝ ΜΕ ΠΟΛΛΑΠΛΗ ΣΚΛΗΡΥΝΣΗ

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Περίληψη

Η σκλήρυνση κατά πλάκας ή πολλαπλή σκλήρυνση (ΠΣ) είναι μια χρόνια απομυελινωτική ασθένεια του κεντρικού νευρικού συστήματος (ΚΝΣ) που χαρακτηρίζεται από μια ευρέως μεταβλητή κλινική εκδήλωση και πορεία. Παρόλο που η σκλήρυνση κατά πλάκας θεωρείται γενικά ασθένεια της λευκής ουσίας, η παθολογία επεκτείνεται και μπορεί να ανιχνευθεί επίσης και στη φαιά ουσία. Η απώλεια των νευρώνων και η νευροεκφύλιση εμφανίζονται νωρίς και μπορεί να οδηγήσουν σε μόνιμη νευρολογική και γνωσιακή εξασθένηση. Η Απεικόνιση Μαγνητικού Συντονισμού (MRI) προσφέρει εξαιρετικές ανατομικές - μορφολογικές πληροφορίες και από το 2001, η MRI ενσωματώθηκε στη διαγνωστική διερεύνηση ασθενών με κλινική υποψία ΠΣ (1). Οι αλλαγές στον όγκο του εγκεφάλου, που ανιχνεύονται από τα πρώτα στάδια της ΠΣ και προχωρούν καθ 'όλη τη διάρκεια της ασθένειας, μπορεί να προσφέρουν ένα μετρήσιμο ποσοτικά μέγεθος της νευροεκφύλισης και της προϊούσας ιστικής βλάβης. Η ποιοτική (δηλ. οπτική) ερμηνεία των μορφολογικών εικόνων του εγκεφάλου μπορεί να ανιχνεύσει μόνο μακροσκοπικές αλλαγές και το επίπεδο συμφωνίας μεταξύ διαφορετικών παρατηρητών φαίνεται χαμηλό. Από την άλλη πλευρά, οι προηγμένες τεχνικές MRI –όπως η ποσοτική αξιολόγηση μέσω της ογκομέτρησης του εγκεφάλου με βάση την MRI– που αναπτύχθηκαν τα τελευταία χρόνια μέσω των εξελίξεων στην υπολογιστική τεχνολογία έχουν μεγαλύτερη ευαισθησία από τη συμβατική MRI και επιτρέπουν την ποσοτική αξιολόγηση ειδικών μορφολογικών ακολουθιών του εγκεφάλου. Δεδομένου ότι η χειροκίνητη αναγνώριση και μέτρηση των βλαβών της ΠΣ στη MRI μπορεί να είναι εξαιρετικά χρονοβόρα και υποκειμενική, έχουν προταθεί πολλές ημι-αυτόματες και αυτοματοποιημένες μέθοδοι για τον προσδιορισμό του συνολικού όγκου του εγκεφάλου, πολλαπλών ξεχωριστών δομών αυτού και του «φορτίου των βλαβών» της ΠΣ. Παρ' όλα αυτά, αν και αυτές οι μέθοδοι φαίνονται ευαίσθητες και αναπαραγώγιμες, ο ρόλος τους θα πρέπει να είναι κυρίως υποστηρικτικός σε αυτόν της ποιοτικής-οπτικής αξιολόγησης των μορφολογικών εικόνων του εγκεφάλου, καθώς μεταβλητοί παράγοντες (π.χ. ψευδοατροφία) μπορούν να έχουν μια συγκεχυμένη επίδραση στις μετρήσεις.

Μετά από μια σύντομη επισκόπηση των διάφορων ογκομετρικών τεχνικών MRI, αυτή η ανασκόπηση θα αξιολογήσει τις κλινικές χρήσεις της ποσοτικής ογκομετρικής αξιολόγησης του εγκεφάλου μέσω ειδικών μεθόδων MRI στη διάγνωση, την παρακολούθηση, αλλά και την αξιολόγηση των θεραπευτικών αποτελεσμάτων σε ασθενείς με ΠΣ.

Λέξεις ευρετηρίου: Απεικόνιση Μαγνητικού Συντονισμού (MRI) εγκεφάλου, ογκομέτρηση (volumetry), Ακτινολογικά Μεμονωμένο Σύνδρομο (RIS), Κλινικά Μεμονωμένο Σύνδρομο (CIS), πολλαπλή σκλήρυνση (MS)

Introduction

Imaging is broadly used in the diagnosis and monitoring of neurological diseases, including MS. MRI has become the cornerstone for the diagnosis, follow-up and management of numerous neurological and psychiatric conditions. In current clinical practice though, these assessments are based on the visual inspection of MR images by experts, who are responsible for the initial diagnosis and for the interpretation of follow-up examinations.

Lately, within the scientific literature, we find an increasing interest in the use of quantitative medical imaging biomarkers, i.e., relevant numerical values that can be extracted with advanced image processing techniques from 2D or 3D image data sets. Many imaging biomarkers, such as volumetric assessment of brain structures, have shown excellent sensitivity and specificity in the diagnosis and prognosis of various neurological diseases, including MS.

The aim of this review is to enlighten the usefulness of quantitative volumetric evaluation of MS patients in the everyday clinical routine for the initial diagnosis, as well as for the follow-up and even in treatment modification, if needed.

Background

MS is a chronic autoimmune, inflammatory and demyelinating disease of the CNS characterized by a widely variable clinical manifestation and course. MS commonly affects young adults and causes changes in the morphology and structure of the brain, leading to disability and cognitive impairment. Although in some cases MS may not directly affect the patients' life expectancy, it has a significant impact on their quality of life.

Establishing the prognosis for MS early in the disease course is important for selecting the appropriate treatment and for determining, throughout the course of disease, when the therapeutic approach should be modified. Nonetheless, our ability to predict how a patient's disease will evolve is still limited. For many years, we have known that specific clinical features of MS are associated with a more benign course (e.g., female gender, clinical onset before the age of 40 years, few early relapses, few early fixed deficits, initial involvement of only sensory systems), but these clinical features offer limited help in decision making since they have a low prognostic value. Throughout the years there have been various attempts to develop paraclinical tests, that could reinforce this clinical prognostic information. Among these, the use of MRI markers, including gadolinium enhancing lesions, new T2 lesions, volume of T2 and/or T1 lesion burden, brain atrophy (either whole brain or separately for grey and white matter), spinal cord atrophy, cortical connectivity, and chemical composition of the normal appearing white matter (NAWM) have been investigated.

Conventional MRI is an excellent, non-invasive imaging technique, very sensitive in detecting brain multifocal WM damage, but it is often not satisfactory in detecting evidence of subtle and widespread abnormalities in the so-called NAWM [2, 3] and in the GM [(4, 5]. The most common MRI protocols (Hashemi et al., 2012) used in discerning MS lesions are T1-weighted (T1-W), T2-weighted (T2-W), PD-weighted (PD-W) and fluid attenuated inversion recovery T2 (T2-FLAIR) sequences. Lesions (also known as "plaques") can be visualized with several MRI sequences: (1) on *T1-WI*: chronic lesions with axonal destruction and irreversible damage appear as dark spots ("black holes"), compared to the surrounding WM tissue intensities; (2) on *gadolinium - enhanced T1 - WI*: "active" inflammatory lesions that enhance indicate breakdown of the blood–brain barrier and ongoing disease activity, since only new lesions (under 6 weeks old) enhance; (3) on *T2-WI, FLAIR and PD-WI*: lesions appear as hyperintense spots compared to the surrounding brain parenchyma.

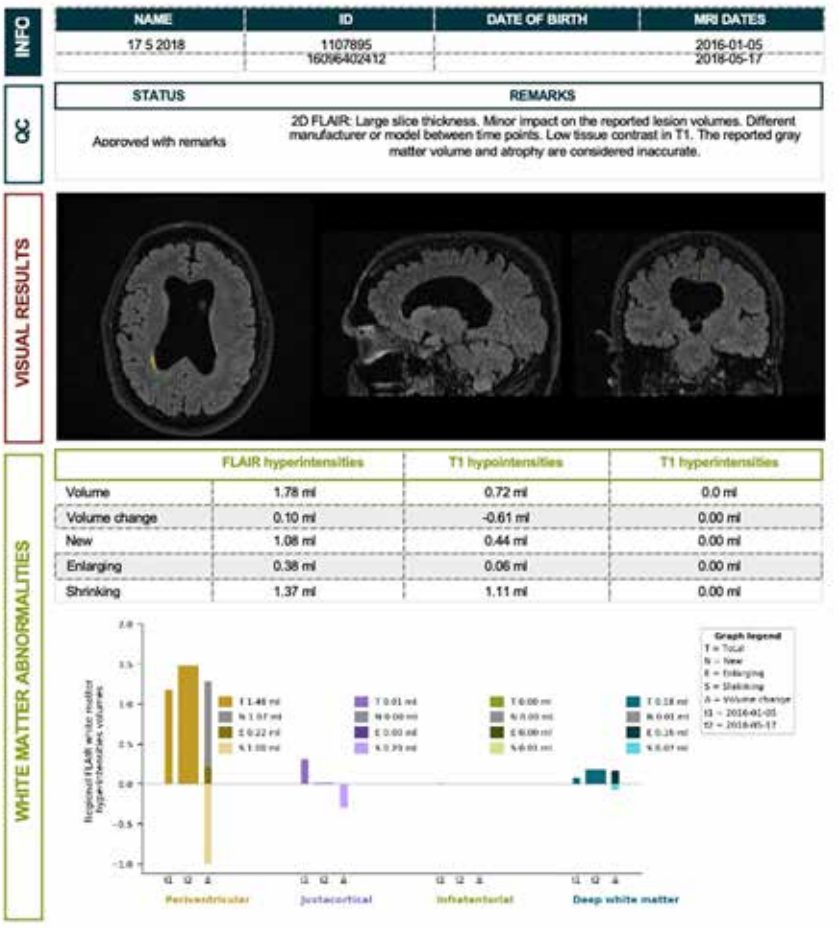
Double inversion recovery (DIR) is an additional MRI sequence that can increase the conspicuity of WM plaques on T2 -WI. It is an inversion recovery MRI pulse sequence that uses two different inversion pulses selected to suppress signal from CSF (e.g. TI_1 = 2000-3000 msec before readout) and from WM (e.g. TI_2 = 450 msec before readout). It is useful in the estimation of lesion load, in the differentiation of juxtacortical from mixed GM-WM plaques, and in the detection of infratentorial or spinal cord lesions. Another MRI sequence helpful in the revelation of spinal cord lesions is T1-W Phase-sensitive IR (PSIR) [6] although it is somewhat tricky to implement since it suffers from phase error artifacts and long scanning times.

The "lesion load", defined as the lesions' total volume in the brain, is one of the key biomarkers in MS. Usually, a distinction is made between T2 lesions (i.e., lesions that appear hyperintense on T2-WI or FLAIR images), T1 lesions (i.e., lesions that appear hypointense on T1-WI, "black holes") and contrast-enhancing lesions (Figure 1a).

Apart from the lesion load, brain volumetry [7] and, more precisely, cerebral atrophy [8] and, specifically, GM atrophy [9] are currently considered to be important biomarkers, that seem to have a positive correlation with the speed of disease progression. Consequently, not only the detection of lesions, but the quantification of brain volumes and atrophy rates is crucial in the management of patients with MS (Figure 1b).

Widespread application of MRI biomarkers is hindered by issues such as non-standardized imaging protocols, imaging artifacts, lack of normative data and manual segmentations to interpret values in

Figure 1a. Longitudinal follow up report (**Icometrix ®)** of a 57-year-old male patient with RRMS**.** The volume of FLAIR and T1-W lesions is estimated. The lesions are classified into N = new, E = enlarging and S = shrinking and their volume is presented in a chart to facilitate their quantitative evaluation and show disease progression



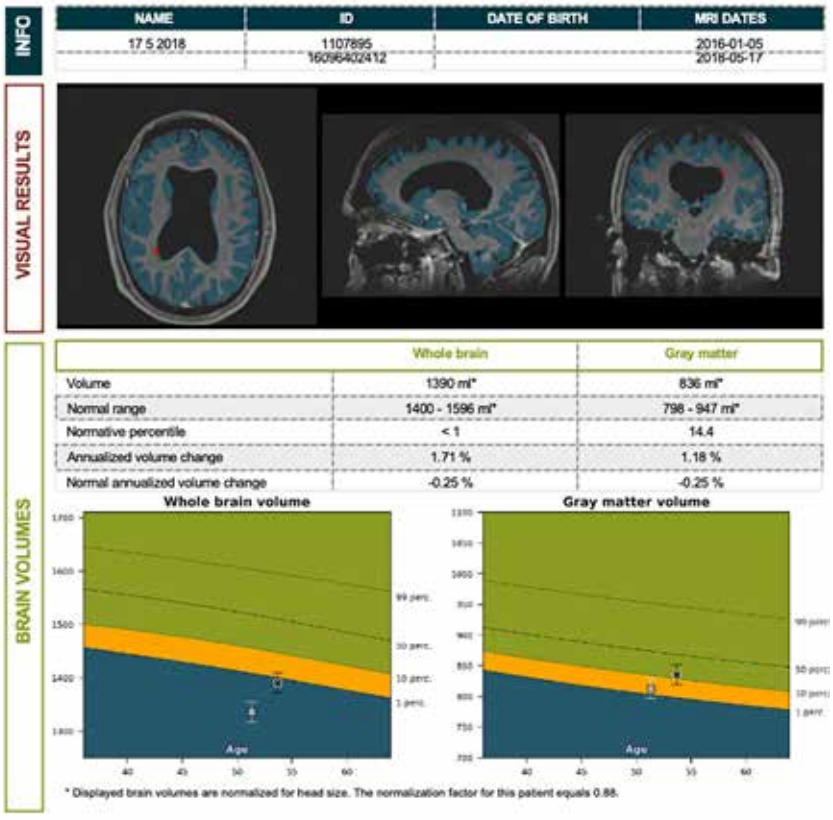
clinical practice. In order to mitigate such issues, the MAGNIMS study group published guidelines for the use of MRI in MS diagnosis [10], as well as recommendations to improve imaging and analysis of brain lesion load and atrophy in longitudinal MS studies [10, 11]. The recommended brain MRI sequences are 3D FLAIR, pre- and post-gadolinium 3D T1-W, axial T2-W and/or PD-W, obtained with a minimum MRI field strength of 1.5 T and near-isotropic spatial resolution. It should be noted that total head coverage should include the entire brain and brainstem.

This article offers a brief review of the different volumetric MRI techniques and discusses the clinical relevance of MRI-derived brain volumetry in the diagnosis and follow-up of patients with MS.

Brain Volume

The volume of the whole brain and of brain structures can be calculated through brain segmentation techniques. Brain segmentation relies on high-contrast borders between brain tissue and cerebral spinal fluid (CSF) (Figure 2a). Standard high-resolution 3D acquisitions (that can be acquired in less than 10 minutes) decrease partial volume effects and yield good CSF/brain and GM/WM contrast, allowing imaging and measurement of small regional structures. Firstly, "brain extraction" is performed to ensure that only brain tissue is conveyed to the segmentation pathway. Various brain extraction methods, such as the brain extraction tool (BET) [12] and the brain surface extractor (BSE) [13] are available and their approaches vary including morphological and geometrical techniques, as well as image processing and modelling functions (hole filling, surface modelling, edge detection, intensity thresholding, atlas matching, etc.). Once brain extraction is completed, the process of brain segmentation begins. This process is typically based on a probabilistic modeling of voxel intensities that takes advantage of the fact that different tissues have different MRI characteristics. Literature provides an excellent overview of brain segmentation

Figure 1b. Automated MRI Brain volumetry system report by **Icometrix®** of the same patient, showing cerebral and GM atrophy; important biomarkers, that seem to have a positive correlation with the speed of disease progression



methods [14] that include FSL FAST [15], SIENAX [16[and FreeSurfer [17]. To give an example, SIENAX [18] (Structural Image Evaluation, using Normalization of Atrophy-Cross-Sectional) uses a fully automated algorithm to quantify the volume of whole brain, GM and WM. An automated BET is used to segment brain from non-brain tissue and appraise the outer skull surface. The brain images are registered to a stereotactic space to perform normalizing for head size and then a brain mask is applied to exclude extracerebral tissue. SIENAX can segment the extracted brain into GM, WM, and CSF with great accuracy and low mean absolute error of volume measurements [18].

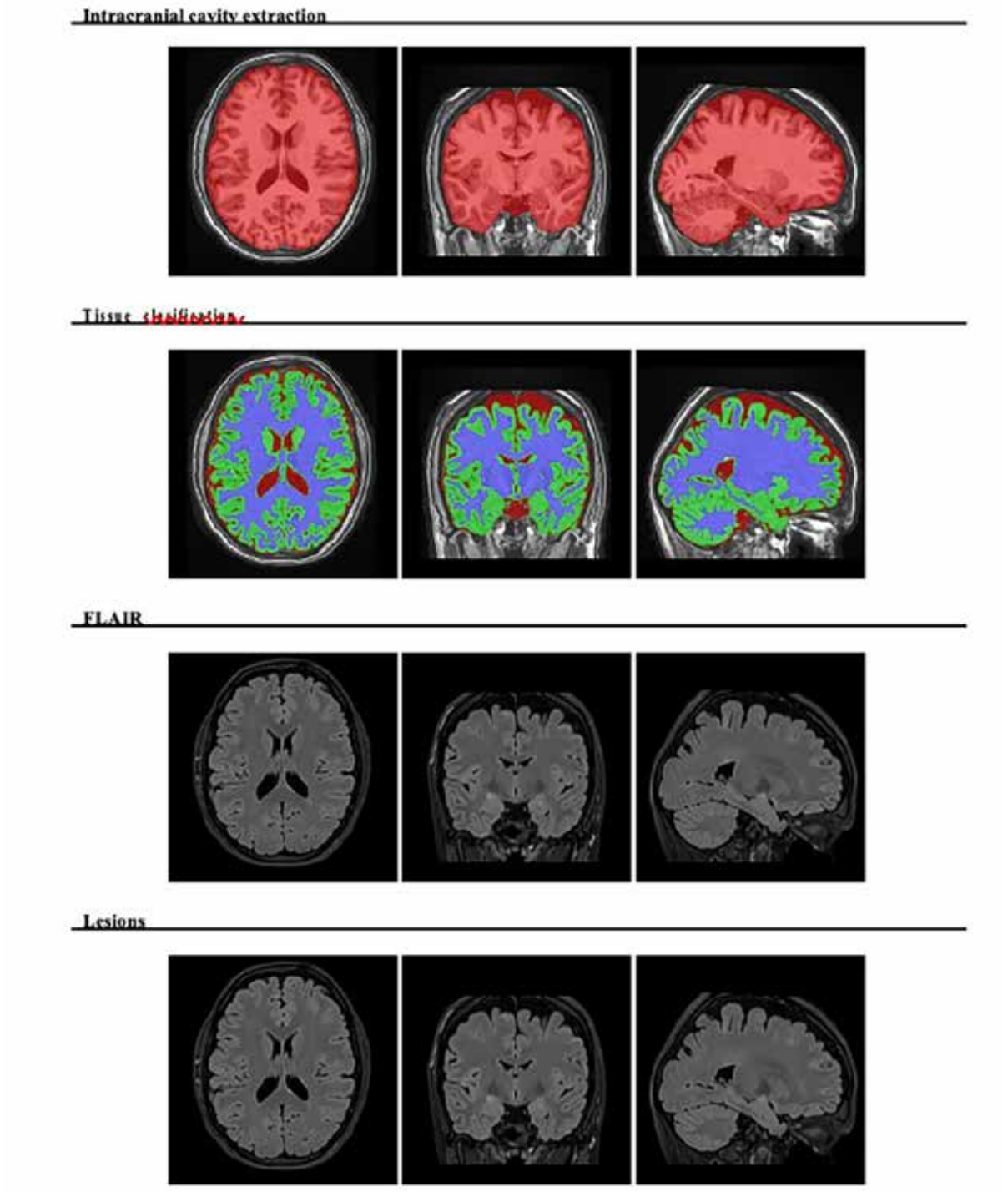
Lesion Detection and Volume Estimation

Automatic lesion segmentation methods can be divided into supervised and unsupervised classification methods. The first group requires a prototypical training dataset in order to build a model that will then be used on new patients for lesion segmentation. Although excellent results can be obtained with the proposed variants of this group [19-22] building a training dataset that incorporates MS lesions of all possible shapes and intensities that are heterogeneously distributed in the WM is difficult. Moreover, it is difficult to preprocess new images –acquired on different scanners than the one used for the training dataset– in order to match the characteristics of the training dataset. The second group includes methods that are mainly based on stochastic modelling of voxel intensity distribution. These perform brain segmentation into GM, WM and CSF (with or without lesion detection) and often depend on post-processing approaches for lesion segmentation. The assumptions made for the lesion segmentation have a great effect on the results. For example, LST [23] and MSmetrix [24] detect FLAIR-hyperintense outliers, which are further characterized as lesions according to their spatial probability of being in the WM, where the WM segmentation is basically derived from T1-WI segmentation. LesionTOADS [25] on the other hand, combines information from different MR sequences in order to synchronously segment lesions and brain structures. In the meantime, the segmented lesions are confined to typical locations by using maps from the boundaries of structures such as CSF (Figure 2).

Longitudinal Biomarkers

Brain volume measurements derived from a single scan are often hard to interpret because there is a

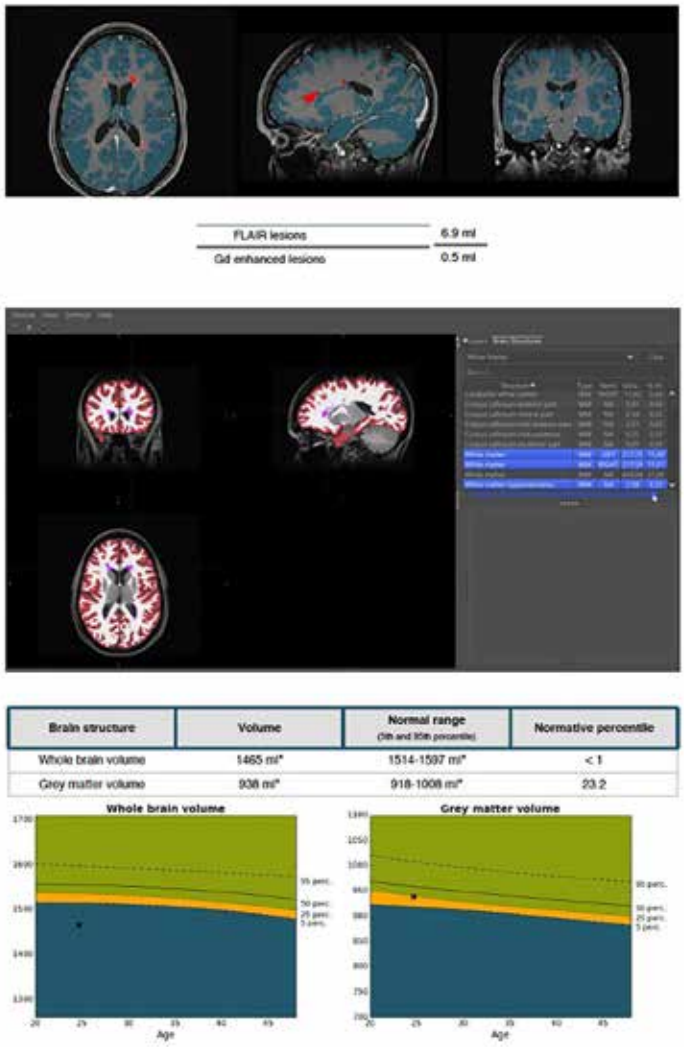
Figure 2. Brain segmentation **(volBrain)** after brain extraction in a 27-year-old female patient with no clinical history of demyelinating attacks or other alternative causes for WM lesions (such as vascular, infectious or toxic) presented with dizziness



broad normal variability. Since small volume changes are apt to being concealed by the interindividual variability in absolute brain volumes, normalization to intracranial volume should be carried out. Longitudinal measurements allow for a more accurate monitoring of the disease progression by identifying the extent of true interindividual differences (Figures 1, 3).

Diffuse atrophy from consecutive scans can be assessed by image subtraction, as less errors occur when we directly quantify the volume change. To do so, however, serial images must be positionally registered (spatially matched). Longitudinal methods typically try to match two MRI scan registration techniques and directly extract small changes in

Figure 3.
(**3a**) Initial MRI based Volumetry report (**Icometrix®**) of a 24-year-old female patient identifies FLAIR lesions with a volume of 6.9 ml and "active" inflammatory (Gd enhanced) lesions with a volume of 0.5 ml, "black holes" volume of 3.08 ml (**Imagilys, SurferMagix®)**. As far as brain atrophy (<1th normative percentile – matched for her gender and age) the report (**Icometrix®**) shows no GM atrophy and normal thalamic volumes (**Imagilys, SurferMagix®**) (**3b**) (**3c**). After treatment, the report (**Icometrix®**) shows decrease of lesion volume in FLAIR (3.54ml). Their distribution is 2.79 ml periventricular, 0.03 ml juxtacortical, 0.09 ml infratentorial and 0.63 ml involving the deep WM. No "active" (Gd-enhanced) lesions where demonstrated. "Black holes" (chronic lesions with axonal destruction and irreversible damage that appear as dark spots on T1-WI) coexist with a volume of 2.14 ml



brain volume from this process. For example, SIENA (Structural Image Evaluation, Using Normalization, of Atrophy) (18) uses the outer skull surface to restrict the registration of serial images while normalizing to image geometry changes. The brain surface is perceived using a local threshold and smoothness factor, and the percentage brain volume change is based on the displacement of this edge between images, with sub-voxel accuracy. Edge finding is relatively insensitive to changes of intensity in tissues through serial images, making this technique applicable to different acquisitions with great accuracy [18].

In what concerns lesion load, there are many methods that focus on segmenting MS lesions at a single time point. However, according to the review of Lladó et al. [26] there is not yet a single approach that can be used as a standard in everyday clinical practice for the analysis of lesion evolution over time, since quantification of atrophy and lesion load using registration-based techniques such as SIENA may be affected by differences in tissue intensity between consecutive images and incorrectly interpret changes in voxel intensity between a baseline and a repeat image as atrophy of the brain or change in lesion load.

Clinical Course and Imaging Findings

From a clinical point of view, MS starts with a "radiologically isolated syndrome" (RIS) or a "clinically isolated syndrome" (CIS) suggestive of MS. RIS [27] refers to an entity in which brain and /or spine MRI reveals serendipitous WM lesions that fulfill the revised 2017 McDonald Criteria for dissemination in space (DIS) but the patient has no clinical history of demyelinating attacks or other alternative causes for the WM lesions such as vascular, infectious or toxic. RIS has been linked to MS and the prevalence of RIS is known to be increased in healthy relatives of patients with MS. In their study Gabelic et al [28] found that the prevalence of RIS in the healthy relatives of patients with MS was 2.9% compared to the prevalence of RIS in unrelated healthy controls that was 2.4%. On the other hand, patients are considered to have CIS when they present with their first clinical symptom suggestive of CNS demyelination but they do not fulfill the McDonald criteria for clinically definite MS. CIS patients who have a normal brain MRI at presentation have only a 5 % risk of progression to clinically definite MS in the next 1-5 years [29]. Diversely, RIS and CIS patients with cerebral lesions on MRI at presentation have a substantially higher risk, although the risk remains below 50 % when the total lesion volume does not exceed 1.2 ml [29].

After disease onset, MS may take one of the following forms: (1) relapsing-remitting MS (RRMS), which is characterized by acute attacks followed by periods of remission; (2) primary progressive MS (PPMS) and (3) secondary progressive MS (SPMS), both of which are predominantly characterized by progressive accumulation of disability, but may also present concomitant clinical and/or MRI activity [30]. MR studies have demonstrated the existence of lesions and the development of brain atrophy in all MS subtypes [31-33].

Clinically the progression of MS is characterized by both motor and cognitive deterioration [34, 35]. Pathological changes in the NAWM have a better correlation with progressive cognitive deficits than with visual, sensory and motor symptoms [9, 36]. Brain atrophy, defined as the diminution of brain volume over time, is considered one of the characteristic consequences of MS even though its pathophysiological mechanisms remain unclear [37]. Patients with MS show a higher rate of brain volume loss compared to healthy controls, that is, 0.5-1 % per year in MS patients versus 0.1-0.3 % per year in age-matched healthy controls [38].

It has been reported that 60-80% of patients with CIS suggestive of MS (e.g., optic neuritis) that undergo MRI evaluation of their brain that detect lesions develop clinically definite MS in the following years [39]. Notably, previous studies have demonstrated greater ventricular enlargement within one year in people who develop MS compared to those who remain stable (+0.5 to 0.8 cm^3 /year compared to -0.1 to +0.06 cm^3 /year) [18]. A three-year follow-up study of 58 patients with CIS observed a 17.9% increase in the ventricular volume in 31 patients who developed MS, while only a 2.3% increase was found in 27 patients who remained stable [40]. Statistical Parametric Mapping (SPM) was used to analyze BPF, WM lesion load and GM lesion load changes in these 58 patients, showing significant decreases in both MS (n = 31) and CIS (n = 27) patients (-1.4% and -0.6% in BPF and -3.3% and -1.1% in GMF, respectively). The decreases were greater in the MS group, with the later also showing a weak but statistically significant increase (1.3%) in WM lesion load.

Okuda et al. [41] in their multicenter study of 451 patients with RIS, found that approximately two-thirds of them developed new lesions on longitudinal MR imaging, while one-third of them developed CIS within 5 years of the index MR imaging.

Various studies have shown that brain volume is notably reduced in RRMS patients when compared to age-matched controls [42-44]. The majority of longitudinal studies –even when including subjects in the earliest stages of MS with no significant disability– have estimated atrophy rates of around -0.5 to -0.8% /year. A study of the mean atrophy rate in 34 subjects, estimated using SIENA, found a 0.7% /year (SD = 0.9) decrease in brain volume over an interval of one year [45].

It is widely accepted that atrophy occurs from the earliest stages of MS and continues throughout the disease course, but it is not yet established if there are differences in the development of atrophy between PPMS and RRMS, or if the atrophy rate changes during the disease course. One study suggested that atrophy is confined to the supratentorial brain during the RR stage, but extends to the brain and spinal cord during the SP phase [42]. A SPM analysis of brain atrophy measured by SIENA was used to explore the evolution of brain atrophy in MS patients according to their subtype (RRMS, SPMS, and PPMS) [46], and the results suggested that while ventricular enlargement was predominant in RRMS, cortical atrophy was the dominant feature in PPMS.

Cross-sectional correlations between normalized brain volume (NBV) and disease duration [43, 47, 48] indicate that brain atrophy is progressive. In a longitudinal analysis of 27 RRMS and 9 SPMS patients brain atrophy was greater in SPMS patients who had a longer disease duration, than in RRMS patients [47]. Another longitudinal study indicated higher annual atrophy rates in RRMS and SPMS compared to PPMS, even though the PPMS patients had a longer disease duration. On the other hand, similar annual atrophy rates were found in PPMS and RRMS patients [49], with reduced rates in SPMS; however,

the differences were not statistically significant and the analysis was conducted on a moderately small number of subjects. Whether atrophy rates decelerate or accelerate during the course of progressive disease is yet to be determined. An analysis of 100 PPMS subjects indicated that the degree of atrophy over the first year did not correlate with that over the second year [50]. However, an ensuing five-year follow-up study indicated a relatively consistent atrophy rate within individuals [51]. For the temporal dynamics of atrophy in MS to be clarified, studies of patients over longer periods are necessary.

It is worth emphasizing, that although focal demyelination in the cerebral WM was until recently considered the pathological hallmark of MS, we now know that in addition to WM lesions MS is also characterized by GM lesions [52]. Brain atrophy in MS is nowadays considered a global process related to both GM and WM pathology. Actually, MRI- based volumetric data have shown that GM atrophy (especially that of thalamus) has a better correlation with physical and cognitive disability than WM atrophy and T1 and T2 lesions do [9, 53, 54]. Morgan et al. examined, and confirmed, the unwritten "rule of five", according to which five new lesions compared to the baseline MRI scan are correlated to a higher risk of ensuing relapses [55]. Sormani et al. followed-up 58 patients with RRMS for a 10-year period and their study confirmed the long-term clinical relevance of lesion load since it showed that the escalation of clinical disability measured by the Expanded Disability Status Scale (EDSS) correlated with the increase of T1 lesion load [56]. Their study showed an annual volume increase of (+0.25 ± 0.5) cm^3 for T2-W lesions and of (+0.20 ± 0.31) cm^3 for T1-W lesions [56]. In the analysis, EDSS worsening over 10 years correlated best with the combination of baseline T1-W lesion number and the increase of T1-W lesion load ($R = 0.61$, $p < 0.001$). Fisniku et al. evaluated the longitudinal relationships between the MRI lesions and clinical course of 107 MS patients over a period of 20 years. They found a lesion load increase of 0.80 cm^3/year in those with RRMS but of 2.89 cm^3/year in those with SPMS [57]. In addition, a multicentre 10-year follow-up study by Popescu et al. showed that brain atrophy and lesion load have a complementary predictive value for disease progression since they correlate with long-term disability in MS [58].

The correlation between cortical lesion load and cognitive impairment has been investigated with conflicting results. Nelson et al. in a study that combined two different MRI techniques, DIR and T1-W phase-sensitive inversion recovery (PSIR), showed that intracortical and mixed lesions play a more important role than juxtacortical lesions and measures of atrophy in cognitive impairment [59]. A longitudinal study of 13 MS patients showed that cortical lesions have a tendency to increase over time and this is associated with a cognitive decline. More specifically, the authors found a significant correlation between the hippocampal lesion load and the location learning test score (LLT), while investigating visuospatial memory [60]. Mike et al. [61] found a similar correlation between cortical lesion load and WM volume damage. They also found that cortical lesions were a good predictor of verbal learning and memory assessed by California verbal learning test (CVLT-II). Calabrese et al. [62] demonstrated that cortical lesions, GM damage volume, and age are good predictors of cognitive impairment, showing also a better correlation of cognitive impairment with cortical lesion load, than with WM damage in MS patients. However, Papadopoulou et al. [63] did not confirm these findings, as their study demonstrated that WM lesion load plays a more important role in the development of cognitive impairment when compared to cortical lesion load.

Treatment monitoring

It is widely accepted that MRI has an indispensable role in the monitoring of disease progression and of therapeutic efficacy [64, 65]. Newly developed treatments, especially those prescribed in the early stages of MS, focus not only on treating the symptoms but on modifying the natural course of the disease. Because of their serious side effects many of these disease-modifying therapies are not prescribed as first-line treatments. Even though imaging criteria for switching from one treatment to another are still under consideration, MRI-based monitoring of therapeutic effects becomes more and more crucial in clinical trials and clinical practice, a fact that increases the need for standardization of MRI-derived metrics [64-66].

Currently, in patients with a clinically active disease (showing relapses and disability progression), who develop at least three active MRI lesions, a change in treatment is recommended [66]. During the course of disease-modifying therapy, new or enlarging lesions should be monitored with MRI at regular intervals, ranging between 3, 6 and 12 months, based on individual clinical assessment and in line with national and international guidelines [67-70]. The development of one or more gadolinium-enhancing lesions on a 6- or 12-month follow-up scan, or two or more new or enlarging lesions on a 12-month follow-up scan, should trigger consideration of treatment modification; however, in cases of isolated MRI-activity (i.e., without evidence of clinical activity) individualized risk/benefit assessment, which besides MRI findings takes into account the patient's history and clinical status, is warranted to guide decisions on potential treatment modification [67, 70, 71].

Clinical trials indicate that the use of lesion load and brain atrophy as endpoints is more efficient than the Expanded Disability Status Scale (EDSS) [54], since it decreases the required sample size for the demonstration of statistically significant therapeutic effects in placebo-controlled trials. Moreover, in clinical practice, MRI indices may facilitate early detection of subclinical disease activity, and instigate timely modification of disease-modifying treatments (DMTs) to avert clinical relapses or EDSS progression.

There is no consensus on whether whole-brain atrophy should be used as the point of reference for the evaluation of therapeutic efficacy after the first year of treatment (38).The main hampering factor is that whole-brain atrophy after 1 year of treatment might be an inaccurate parameter, due to the occurrence of pseudo-atrophy, with the latter representing an early decrease of brain volume due to the diminution of inflammation [72]. Pseudo-atrophy seems to affect WM more than GM [38] and may persist for more than 2 years after treatment initiation. Therefore, measuring GM atrophy, instead of whole-brain atrophy, might be more useful.

Some clinical trials have shown that brain volume loss (or GM volume loss) is a good prognostic indicator for the natural course of the disease [72]. However, in clinical trials where DMTs were used, the results were contradictory. The inconsistency between the acquired results could be attributed to various factors, such as differences in the action mechanism of DMTs, patient populations, applied MRI protocols and software applications used for the analysis.

It is obvious that if brain atrophy is to be used as a representative marker of axonal loss in clinical trials, reliable detection of atrophy over short intervals would probably ameliorate trial efficiency and reduce costs. However, the effect of measurement errors on brain atrophy quantification is increased over short intervals; therefore larger subject numbers are needed for credible results to be obtained. For example, in a study of 30 RRMS patients, no significant brain atrophy was found over three months [73].

Agents such as interferon and glatiramer acetate are considered efficacious in lowering relapse rates and lesion formation, but it is still uncertain whether they actually slow down neurodegeneration and disability progression. Although brain atrophy cannot be used to directly monitor neurodegeneration, it has been used as a representative marker and outcome measure in several interferon-β trials in MS (Table 1). Brain atrophy seems to progress despite treatment with these alleged disease-modifying drugs; however, the results of longer follow-up periods in studies by Paolillo et al [74] and Turner et al [75] suggest that either therapeutic action is delayed or a beneficial effect from baseline becomes apparent only in later atrophy measures. It is becoming increasingly important to develop neuroprotective strategies, and brain atrophy rates are likely to be included as outcome measures in such strategies. The acquisition protocols and atrophy measurement techniques used in clinical trials must be highly reproducible and robust, since the data will be acquired over multiple sites (Table 1).

Relationship Between Lesions and Brain Atrophy

It is widely accepted that focal inflammation can eventually cause brain atrophy; nonetheless, attempts to correlate brain atrophy with lesion measures have produced diverse results. In the literature we find studies of CIS and MS that show an association between brain volumes and T1-hypointense lesions [40, 76, 77], T2-hyperintense lesions [40, 42, 44, 76-78], and gadolinium (Gd) -enhancing lesions [79] and others that do not [47, 72, 76]. It is evident that to evaluate such a relationship consecutive data are needed.

CIS patients that had T1-, T2-, or Gd-enhancing lesions at baseline showed greater ventricular enlargement over a period of one year compared to patients who had no lesions on baseline imaging [80]. In the existing literature we find many studies that show a relationship between brain atrophy and change in lesion volumes over the same period of time [48, 50, 81]; however, we also find studies that indicate no correlation [82]. Studies that included therapeutic trials indicated that the number and volume of Gd-enhancing lesions discerned during the early study periods were highly correlated with brain atrophy developed over periods of 18 months to three years in CIS and MS patients [79, 82-84]. This is probably due to the fact that focal inflammation seems to have a delayed effect on neuroaxonal degeneration and ensuing atrophy. Even though many studies have shown mixed results about the existing relationship between lesions at baseline and subsequent atrophy [81, 85, 86], findings from studies that included a longer follow-up period enhance the belief that atrophy results from earlier inflammation. Chard et al. followed 28 RRMS patients for 14 years after their initial onset of symptoms and discovered that the alterations in lesion load during the first five years was more closely correlated to brain atrophy at 14 years than later alterations in lesion load [87].

In order to explain the fact that some studies have found no correlation between lesions and atrophy one may consider the presence of small lesion loads, the fact that lesion activity may lead to variable degrees of axonal damage, the presence of inflammation causing edema, or the fact that GM lesions could have a greater effect on following atrophy than WM lesions. Additionally, there is evidence that factors not related to lesion formation play a part in the progression of atrophy. It is worth noting that Inglese et al. showed in their study that their attempt to suppress the inflammation with autologous hematopoietic stem cell transplantation this did have a significant

Table 1. Indicative Therapeutic Trials of Interferon β That Have Used Brain Atrophy as an Outcome Measure

Subjects	Study Design	Atrophy measure	Comments	Reference number
263 CIS	123 treated subjects (22 μg) with 2 year data. 117 assigned placebo with 2 year data.MRI at baseline, 1 year and 2 years.	SIENA	Brain atrophy rate 1.18% reduction (SD 1.51) in treated and 1.68% reduction (SD 1.99) in placebo over a 2 years period.	(91)
			Significant treatment effect detected at 2 years.	
			31% of treated and 47% of placebo developed MS.	
			Median 2 year atrophy rate 1.63% and 0.97% reduction in those developing MS compared to those who did not ($P = 0.046$)	
386 RRMS	189 subjects treated for 3 years (30 μg).	Brain parenchymal fraction	Brain atrophy greater during the first year than the following 2 years.	(92)
	197 subjects treated for 3 years (60 μg).MRI at baseline, 1 year, 2 years and 3 years.		68% of the atrophy occurring during the first year occurred during first 4 months of treatment.	
	138 subjects had MRI at 3 months, and 2 months before treatment, and months 4, 5, 6, 10, and 11.		Annual atrophy rates were evaluated -0.33% (95% CI -1.50 to -0.62) during the treatment phase and -1.06% (95% CI -0.39 to -0.27) during the pretreatment phase.	
140 RRMS	68 subjects treated for 2 years.	Brain parenchymal fraction	Similar rate of brain atrophy during year 1 was found in both treated and placebo groups.	(48)
	72 assigned placebo.		Brain atrophy rate was significantly lowered during year 2 in the treated group.	
	MRI at baseline, 1 year and 2 years.			
106 RRMS	106 subjects treated for 2 years. MRI at baseline, 2 years and 8 years.	Brain parenchymal fraction	Significant brain atrophy over 8 years.	(93)

and sustained effect on Gd enhancement and T2 lesion formation but did in the long term it did not hinder atrophy [88].

We should also contemplate the fact that lesions may affect both segmentation- and registration-based brain atrophy measurements. For example, it is possible for T1-hypointense lesions to be incorrectly classified as CSF or for GM lesions to cause indistinct signal intensity changes and affect segmentation. However, Sharma et al. analysed 10 MS patients with high T1 lesion loads and found that lesion misclassification had a frivolous effect on brain volume measurements by SPM [89]. Likewise, in the literature we find studies that show no significant difference in tissue volumes estimated with SPM between images with simulated WM lesions and those without [90]. We may assume that brain volumes calculated with SPM segmentations are relatively insensitive to WM lesions, and therefore measurement of GM atrophy may provide a more thorough assessment of neurodegeneration in MS, not affected by alterations in tissue volume due to inflammation.

Conclusion

MS is a chronic demyelinating disease of the cen-

tral nervous system that leads to disability and cognitive impairment. Conventional MRI is considered the modality of choice for diagnosing and following up patients with MS. Manual detection of MS lesions in the MRI images is a time-consuming and subjective process. Newly developed MRI methods and software applications can assist in the quantitative evaluation of MS. Both segmentation- and registration-MRI based methods may be used to measure brain volume. Brain volume loss and in particular GM volume loss occurs early and progresses throughout the course of MS; it is considered one of the most valid prognostic parameters of subsequent disability progression in patients with MS. Nonetheless, serious considerations should be made before widely implementing it in clinical practice, mainly because despite recent progress, there is not yet a robust enough automated lesion segmentation approach. It seems that there is still room for improvement; new algorithms and advances in MRI acquisition protocols will unequivocally assist neuroradiologists in the early diagnosis, follow up and assessment of treatment effects.

Conflicts of Interest Statement

The authors declare no conflicts of interest.

Consent for publication

All patients provided written informed consent for the scientific use of de-identified imaging data.

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