Spatial mapping of T_2 and gadolinium-enhancing T_1 lesion volumes in multiple sclerosis: evidence for distinct mechanisms of lesion genesis?


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Summary

It is generally believed that most T_2-weighted (T_2) lesions in the central white matter of patients with multiple sclerosis begin with a variable period of T_1-weighted (T_1) gadolinium (Gd) enhancement and that T_1 Gd-enhancing and T_2 lesions represent stages of a single pathological process. Lesion probability maps can be used to test this hypothesis by providing a quantitative description of the spatial distribution of these two types of lesions across a patient population. The simplest prediction of this hypothesis would be that the spatial distributions of T_1 Gd-enhancing lesions are identical. We generated T_1 Gd-enhancing and T_2 lesion probability maps from 19 patients with relapsing–remitting multiple sclerosis. There was a significantly higher probability (P < 0.001) for T_2 lesions to be found in the central relative to the peripheral white matter (risk ratio 4.5), although the relative distribution of T_1 Gd-enhancing lesions was not significantly different (P = 0.7) between central and peripheral white matter regions (risk ratio 0.6). Longitudinal data on the same population were used to demonstrate a similar distribution asymmetry between new T_1 Gd-enhancing and new T_2 lesions that developed over the course of 1 year. Alternative hypotheses to explain this observation were tested. We found no spatial difference in the likelihood of development of persistent T_2 lesions following T_1 Gd enhancement. The relative distribution of T_1 Gd-enhancing lesions was shown to be independent of the dose of Gd contrast agent and the frequency of scanning. Our findings suggest that a proportion of the periventricular T_2 lesion volume may arise from mechanisms other than those associated with early breakdown of the blood–brain barrier leading to T_1 Gd enhancement.

Keywords: multiple sclerosis; MRI; neuropathology; plaque; inflammation

Abbreviations: Gd = gadolinium, LPM = lesion probability map; SPAM = statistical parametric anatomical map; SPM = statistical parametric map; T_1 = T_1-weighted; T_2 = T_2-weighted; TE = echo time; TR = repetition time

Introduction

Multiple sclerosis is a chronic CNS disorder characterized by multifocal inflammatory white matter lesions. The underlying pathology is complex and no fully satisfactory animal model is available. Brain MRI has become the single most important tool for understanding the dynamic pathology of this condition (Miller, 1994). T_2-weighted MRI defines lesions with high sensitivity in multiple sclerosis and is used as a measure of disease burden. However, such high sensitivity occurs at the expense of specificity, as T_2 signal changes can reflect areas of oedema, demyelination, gliosis and axonal loss. Poor pathological specificity may in part explain the weak correlation seen between total brain T_2 lesion volumes and disability (Barkhof and Filippi, 1995; Filippi et al., 1995a). Classical pathological studies and more recent imaging studies (Narayanan et al., 1997) emphasize that T_2 lesions have a predilection for particular areas of the CNS. In particular, T_2 lesions cluster around the lateral ventricles and within the corpus callosum. However, with the recognition that
substantial axonal injury may occur secondarily to the inflammatory process, it is not clear that all increases in periventricular T2 lesion volume result entirely from the same pathological processes that give rise to new discrete T2 lesions within the more peripheral white matter regions. It is possible, for example, that periventricular changes may, in part, represent consequences of Wallerian degeneration secondary to distant, peripheral axonal injury.

Areas of gadolinium (Gd) enhancement demonstrated on T1-weighted MRI are believed to reflect underlying blood–brain barrier disruption from active perivascular inflammation (Kermode et al., 1990). Such areas of enhancement are transient, typically lasting <1 month (Miller et al., 1988). Gadolinium-enhanced T1-weighted MRI are therefore used to assess disease activity (Miller et al., 1993). However, although there is correlation with short-term disability (Koudriavtseva et al., 1997), the significance of enhancement for the development of later fixed disability is not established.

The precise temporal and spatial relationships between T2 and T1 Gd-enhancing lesions are not well defined. Careful follow-up of discrete, manually identified lesions in individual patients suggests that discrete new hyperintense T2 lesions arise from areas of previous blood–brain barrier breakdown (Miller et al., 1988; Bastianello et al., 1990; Thompson et al., 1992). However, the time course for these studies has necessarily been limited and manual detection of changes in T2 lesion size in areas of high lesion density, such as the periventricular region, are difficult to identify accurately. A more accurate description of the relationship between T1 Gd-enhancing and T2 lesions might be gained from methods that allow more automated quantitative analysis after pooling of patient data sets.

Evans and colleagues have proposed the use of statistical parametric anatomical maps (SPAMs) to describe brain morphology across large populations in probabilistic terms (Penhume et al., 1996). We have extended the concept of mapping normal brain anatomy to mapping MRI markers of disease in multiple sclerosis. In an initial application of the use of lesion probability maps (LPMs), we defined quantitatively the spatial distribution of T2 lesions in groups of patients with relapsing–remitting and secondary progressive multiple sclerosis (Narayanan et al., 1997). Similar overall lesion distribution with prominent clustering of lesions within the corpus callosum and periventricular white matter was found for the two clinical groups.

Pooling information from large data sets using LPMs may allow patterns of disease activity to be visualized to define features that are less clear in studies of single individuals. The approach may be useful, particularly for studies of relatively low-frequency events such as T1 Gd enhancement or when differences within single patients may be small, such as in detecting subtle changes in confluent periventricular T2 lesions. Using this approach, we set out in the present study to compare the spatial relationships of T1 Gd-enhancing and T2 lesions in a population of patients with relapsing multiple sclerosis in order to test the hypothesis that T1 Gd enhancement invariably precedes the development of all T2 lesions.

**Methods**

The scanning study was approved and conducted according to guidelines of the Central Oxford Regional Ethics Committee (COREC). Nineteen patients with clinically definite relapsing multiple sclerosis who had suffered one or more relapses in the preceding 18 months and had been clinically stable for the previous month were recruited and gave informed consent to participation in the study. Patients were ambulatory (EDSS 1–5.5) and aged between 20 and 55 years. MRI examinations were performed at intervals of 3 months on a 1.5 T GE Signa imager for 1 year. Scout images were taken to confirm the similar positioning of all subjects, and axial scanning was performed parallel to the plane defined by the anterior and posterior commissures. A long repetition time (TR) dual-echo spin echo sequence was obtained [TR 2500 ms, echo time (TE)1 20 ms, TE2 100 ms, 24 × 5 mm contiguous axial slices]. This was followed by pre- and post-Gd (0.1 mmol/kg) short TR spin echo sequences (TR 400 ms, TE 13 ms, 24 × 5 mm contiguous axial slices) with a 5-min delay between contrast injection and scanning. Image segmentation and analysis were performed with software developed at the Montreal Neurological Institute (Collins et al., 1994). Registration to the standard brain was performed with statistical parametric mapping (SPM96) (Friston et al., 1995) using a linear transformation. Nearest neighbour interpolation was used for transformation of all binary lesion masks and trilinear interpolation for grey-scale images, so that registration should not have resulted in any artefactual change in lesion volumes. The data were examined in two principle forms: (i) a baseline cross-sectional comparison of T1 Gd-enhancing and T2 lesion volume distribution, and (ii) a longitudinal comparison of the spatial distribution of new T1 Gd-enhancing and new T2 lesion volumes.

**Cross-sectional T1 Gd-enhancing and T2 lesion probability maps**

Lesions of individual patients were segmented at baseline by a single observer using a manually defined edge thresholding technique. In preliminary studies inter-rater variability was demonstrated to be <8%. The T2-weighted images were segmented using software that allowed the operator to directly toggle between the T2 and co-acquired proton density image to allow more accurate differentiation of periventricular T2 lesions from CSF. The T1 Gd-enhancing lesions were segmented using the same algorithm with the observer blinded to patient identity and the distribution of T2 lesions. Lesions within the cortical grey matter were excluded from the analysis. Patients’ segmented brain images were then warped into a common standard brain space by registering them with the Montreal Neurological Institute average brain (n > 300)
Fig. 1 Lesion probability maps (LPMs) in standard brain space for $T_2$ lesion volumes (A–C) and $T_1$ Gd-enhancing lesion volumes (D–F) for the study population at baseline. The 300 cm$^3$ periventricular mask defining the central and peripheral white matter regions is also shown (G–I). LPMs and the periventricular mask are superimposed on the standard brain. The probability of lesion occurrence at any single point across the study population is given by the pixel intensity at that point. Lesion probability is encoded on a hot metal scale (shown to the left), higher probability being indicated by bright areas. There is prominent periventricular clustering of $T_2$ lesion volumes and a more dispersed and peripheral distribution of $T_1$ Gd-enhancing lesion volume.

coeextensive with the Talairach atlas (Talairach and Tournoux, 1988; Evans et al., 1993). Accuracy of registration with the standard brain was confirmed visually and by demonstrating that the subtracted brain images in standard space approximated to a null image. In general, small degrees of atrophy within the multiple sclerosis patient group did not significantly affect registration with the standard brain template. However, in instances where registration with the standard brain was poor using the automated algorithm, warping was repeated using manually defined starting tag points. All the segmented lesion volumes were then summed in standard space, across the patient group, and normalized to create the lesion probability maps for $T_1$ Gd-enhancing and $T_2$ lesions (Fig. 1). Within these maps, the probability
of finding a lesion in any given voxel is defined by the relative voxel intensity. The intensity of any voxel is directly proportional to the frequency with which lesions occupy that voxel across the study population.

In order to quantify any distribution asymmetry between the periventricular and more peripheral white matter regions for T1 Gd-enhancing and T2 lesion distributions, a central periventricular region was defined by creating a mask in standard brain space. A 300 cm³ mask was created by segmenting and blurring the lateral and third ventricles on the standard brain template (Fig. 1). This mask was applied to each patient’s segmented image volume in standard space and the relative volumes of T1 Gd-enhancing and T2 lesions in the central and the peripheral white matter regions were determined separately. If T2 lesion formation were critically dependent upon prior T1 Gd enhancement, then the spatial distributions of these lesions in the two defined white matter regions should be similar.

To determine the probability of lesion per unit volume within the two defined white matter regions, the volume of white matter, as defined by the mask, within the periventricular and peripheral white matter regions was calculated. This was done using the statistical neuro-anatomical maps generated from 305 MRI volumes (Evans et al., 1993) for white matter, grey matter and CSF. These tissue class probability maps are coextensive with the standard brain space to which the patient image volumes had been registered.

**Longitudinal T1 Gd-enhancing and T2 lesion probability maps**

The cross-sectional LPM analysis does not take into account the fact that T2 lesions are relatively persistent compared with T1 Gd-enhancing lesions. Differences in T1 Gd-enhancing and T2 lesion volume distribution might arise if there were, for example, a relative deficit of new T1 Gd-enhancing lesions in the central white matter region with more chronic disease. To examine this possibility, LPMs were generated from new T1 Gd-enhancing and new T2 lesion volumes identified in follow-up. Cumulative T1 Gd-enhancing lesion maps for each patient over the year were generated by registering T1 scans for each patient at months 3, 6 and 9 with the initial scan from month 0. New T1 Gd-enhancing lesions were segmented on each scan and the segmented image volumes were then summed in native space to produce a cumulative T1 Gd-enhancing lesion map for that patient. The cumulative T1 Gd-enhancing lesion maps from all patients were warped into the standard brain space as previously described, and summed together to produce a cumulative T1 Gd-enhancing LPM for the study population. This map defined the likelihood of any voxel containing new T1 Gd-enhancing lesions across the study group.

A new T2 LPM defining only new T2 lesions over the same period was generated by summing T2 lesion difference maps in standard space. Difference maps define areas of new and resolved T2 lesions and are calculated for each patient in native space (Lee et al., 1998). Each of these was created by registering the final T2 scan (month 12) with the T2 entry scan for time 0. All T2 lesions were then segmented in both image volumes and the segmented image volumes at time 0 were subtracted from that at month 12 to create a T2 difference image. Areas of unchanged T2 lesion cancel, new T2 lesion volume gives a positive intensity and resolved T2 lesion gives a negative intensity. For the principle analysis, areas of T2 lesion volume resolution were removed. The new T2 lesion difference maps for each patient were then warped to the standard brain space and summed to generate a new T2 lesion LPM defining the distribution of new T2 lesions across the study population after 1 year. Relative lesion distributions were measured by calculating the ratios of central to peripheral white matter new T1 Gd-enhancing and new T2 lesion distribution using the previously defined periventricular mask.

**Longitudinal T1 Gd-enhancing/T2 lesion correlations**

To address the possibility that the periventricular white matter might be preferentially susceptible to inflammatory damage leading to T2 lesion, we measured the volume of new T1 Gd-enhancing lesion volume spatially associated with new T2 lesion volume as a fraction of the total new T1 Gd-enhancing lesions in central and peripheral white matter regions, respectively, i.e. the likelihood of new T1 Gd-enhancing lesions forming new T2 lesions in the two regions of interest. This was done by applying the cumulative T1 Gd-enhancing lesion map for each patient to the new T2 lesion map for each patient in standard space and the volume of overlap (i.e. T1 Gd enhancement overlap with new T2 lesions) measured in both the central and the peripheral white matter region using the standard mask. The total volume of new T1 Gd-enhancing lesions and new T2 lesion overlap was also calculated in native space to provide a potentially more accurate overall measure of the likelihood of new T1 Gd enhancement resulting in persistent T2 lesions. Lesion probability maps were also generated for volumes of resolved T2 lesions between baseline and 1-year T2 scans to compare the extent of T2 lesion resolution between peripheral and central white matter regions.

**Cross-sectional single- versus triple-dose T1 Gd-enhancing lesion probability maps**

To address the possibility that any distributional asymmetry found between T2 and Gd-enhancing T1 lesions might reflect sampling error associated with 3-monthly, single-dose, contrast-enhanced T1 scanning, further analysis was performed using data from two cohorts of multiple sclerosis patients. The first analysis was performed to compare the
spatial distribution of Gd-enhancing T1 lesions detected with single-dose Gd contrast with those detected with triple-dose contrast. Cross-sectional data were analysed from 36 patients with relapsing multiple sclerosis undergoing T1 imaging (1.5 T, SE TR = 768 ms, TE = 14 ms, 5 mm contiguous axial slices, field of view 230 mm) performed 5–7 min after single-dose (0.1 mmol/kg) and triple-dose (0.3 mmol/kg) Gd contrast as part of another study (Filippi et al., 1988). All images were collected within 24 h of each other. LPMs were generated for both single- and triple-dose Gd-enhancing T1 lesions using the techniques described above, and lesion distributions were quantified using the periventricular mask.

Longitudinal T1 Gd-enhancing lesion probability maps from weekly scans

Longitudinal data were analysed from five multiple sclerosis patients who had undergone weekly standard-dose Gd contrast T1 imaging for 3 months (1.5 T, TR = 500 ms, TE = 14 ms, 5 mm contiguous axial slice, field of view 240 mm) in order to determine the axial distribution of more transiently enhancing T1 lesions. An LPM was generated for all new T1 Gd-enhancing lesions seen on weekly scans and the periventricular : peripheral lesion volume ratio was measured as before.

Statistics

Null hypotheses were tested using the Wilcoxon signed rank test with a two-sided significance level of 5%.

Results

Clinical descriptions of the patients and quantitative T1 Gd-enhancing and T2 lesion data for the study population are shown in Table 1. Only five patients had no evidence of enhancement at baseline and two patients had no evidence of enhancement at any time during the study.

Cross-sectional T1 Gd-enhancing and T2 lesion distributions

T1 Gd-enhancing and T2 lesion probability maps were created for the study population for scans obtained at the start of the study period (Fig. 1). The T2 LPM showed a high probability for T2 lesions within the corpus callosum and around the body and occipital horns of the lateral ventricles and (to a lesser extent) within the internal capsule. The T1 Gd-enhancing LPM demonstrated a reduced overall intensity relative to that of the T2 LPM, reflecting the much smaller volume of T1 Gd-enhancing lesions present at any time. Qualitative comparison suggested that the spatial distribution of the T1 Gd-enhancing lesions was more peripheral and dispersed than the distribution of T2 lesions.

Quantitative evaluation revealed a median central : peripheral lesion volume ratio for T2 lesions of 2.4 (range 0.8–15), which was significantly greater ($P = 0.001$) than the spatial distribution ratio for T1 Gd-enhancing lesions (0.3, range 0–1.6) (Fig. 2). Two patients were excluded from the analysis: one patient had all T2 lesions in the central region and one patient had all T1 Gd-enhancing lesions in the central region (giving infinite distribution ratios). Only one patient had a greater T2 lesion volume peripherally than centrally (distribution ratio $<1$), while ten patients had a greater T1 Gd-enhancing lesion volume peripherally than centrally (Fig. 2).

The volume of white matter contained within the periventricular mask was 153 cm$^3$, with 289 cm$^3$ of white matter outside the mask. The median increased risk for a T2 lesion expressed per voxel in the central relative to the peripheral white matter region was therefore 4.5. This was significantly greater than unity ($P = 0.001$), confirming that there was a volume-normalized increase in likelihood for T2 lesions to occur within the periventricular white matter compared with the peripheral region. For T1 Gd-enhancing lesions the median probability per voxel of 0.6 for the central white matter compared with the peripheral region was not significantly different from unity ($P = 0.7$), suggesting that there was no substantial tendency for T1 Gd-enhancing lesions to be distributed preferentially in either white matter region.

To confirm that the distributional asymmetry between T1 Gd-enhancing and T2 lesions is independent of the size of periventricular mask chosen, the relative distribution ratio for both types of lesion volumes was calculated for a range of different mask volumes (Fig. 3). The central : peripheral

<table>
<thead>
<tr>
<th>Number of patients (M : F)</th>
<th>19 (4 : 15)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age (range)</td>
<td>38.3 years (28–50)</td>
</tr>
<tr>
<td>Median EDSS (range)</td>
<td>2.5 (2–5.5)</td>
</tr>
<tr>
<td>Median T2 lesion volume at baseline (range)</td>
<td>9.8 cm$^3$ (0.2–61.2 cm$^3$)</td>
</tr>
<tr>
<td>Median T2 lesion volume at 1 year (range)</td>
<td>11.3 cm$^3$ (0.3–71.0 cm$^3$)</td>
</tr>
<tr>
<td>Total net T2 lesion volume change for year</td>
<td>32.1 cm$^3$</td>
</tr>
<tr>
<td>Total new T2 lesion volume change for year</td>
<td>99.1 cm$^3$</td>
</tr>
<tr>
<td>Total number of T1 Gd-enhancing lesions at baseline</td>
<td>57</td>
</tr>
<tr>
<td>*Total number of T1 Gd-enhancing lesions over 1 year</td>
<td>147</td>
</tr>
<tr>
<td>Total volume of T1 Gd-enhancing lesions at baseline</td>
<td>6.3 cm$^3$</td>
</tr>
<tr>
<td>*Total volume of T1 Gd-enhancing lesions over 1 year</td>
<td>18.5 cm$^3$</td>
</tr>
</tbody>
</table>

*Based on 3-monthly scanning.
Fig. 2 Cross-sectional central : peripheral lesion volume ratios for T\textsubscript{1} Gd-enhancing (black columns) and T\textsubscript{2} (open columns) lesion volumes for the study patients at baseline. Five patients had no T\textsubscript{1} Gd-enhancing lesion at entry to the study. A ratio of 1 (intercept) indicates a volume of lesion in the central periventricular white matter equal to that in the more peripheral white matter region. Ratios greater than unity (upwards) indicate greater lesion volume within the central relative to the peripheral white matter region. Ratios less than unity (downwards) indicate greater lesion volume in the peripheral relative to the central white matter region. The marked central white matter distribution bias for T\textsubscript{2} lesion volume is highly significantly ($P < 0.001$) different from the more peripheral T\textsubscript{1} Gd-enhancing lesion volume distribution.

Lesion volume ratio was always greater for T\textsubscript{2} lesions than for T\textsubscript{1} Gd-enhancing lesions for all mask volumes tested. The relative distribution of lesions was tested further by determining the density of T\textsubscript{1} Gd-enhancing and T\textsubscript{2} lesions within a series of successively larger white matter annuli defined using increasing periventricular mask volumes extending out from the central, periventricular white matter (Fig. 4). This analysis confirmed that T\textsubscript{2} lesion density is highest in the intermediate periventricular white matter region and decreases until an approximately constant lesion density is reached in the most peripheral (approximately) 50% of the white matter. In contrast, the T\textsubscript{1} Gd-enhancing lesion density remains approximately constant from central through peripheral white matter regions.

**Longitudinal new T\textsubscript{1} Gd-enhancing and new T\textsubscript{2} lesion distributions**

The LPM for new T\textsubscript{2} lesion volumes (data not shown) was qualitatively similar to the cross-sectional total T\textsubscript{2} lesion volume LPM. There was marked callosal and periventricular clustering of the new T\textsubscript{2} lesion volume. The new T\textsubscript{1} Gd-enhancing LPM again demonstrated a more diffuse and peripheral lesion probability distribution than the new T\textsubscript{2} LPM. A highly significant difference ($P = 0.001$) between the median central : peripheral lesion volume ratio for the new T\textsubscript{2} lesions (2.4, range 0.5–6.3) and the T\textsubscript{1} Gd-enhancing lesions (0.6, range 0–2.2) was found (Fig. 5).

**Longitudinal T\textsubscript{1} Gd-enhancing/T\textsubscript{2} lesion correlations**

The preferential clustering of new T\textsubscript{2} lesion volumes in the central white matter could result from greater persistence of T\textsubscript{2} lesion changes than is found in peripheral white matter. However, the median probability for the association of a T\textsubscript{1} Gd-enhancing lesion with a subsequent, persistent T\textsubscript{2} lesion at 1 year was not significantly different ($P = 0.5$) between central and peripheral white matter regions, although small imprecisions in warping to the standard brain template will have reduced the power of this analysis. Analysis of overlap of new T\textsubscript{1} Gd-enhancing lesion with new persistent T\textsubscript{2} lesion in native space, where subvoxel registration accuracy is typical, demonstrated a median overall likelihood of 19.4% that a T\textsubscript{1} Gd-enhancing lesion would directly result in persistent new T\textsubscript{2} lesions. An alternative explanation for preferential clustering of new T\textsubscript{2} lesion volumes in the central white matter might be that resolution of new lesions is more likely in the peripheral than in the central white matter. Analysis of the distribution of T\textsubscript{2} lesions that had resolved over the course of 1 year revealed a central : peripheral resolved T\textsubscript{2} lesion volume ratio that was not significantly different from unity (median 1.3, range 0.45–30), implying that there was no substantial increase in the tendency for T\textsubscript{2}
Cross-sectional single- versus triple-dose $T_1$ Gd-enhancing lesion probability maps

To determine whether the distribution asymmetry arises from differences in the relative magnitude of blood–brain barrier breakdown in central versus peripheral white matter regions, we compared $T_1$ Gd-enhancing LPMs from both single- and triple-dose scans. One hundred and five lesions were detected on single-dose contrast-enhanced $T_1$ scans compared with 205 lesions detected using triple-dose contrast based on scans collected in an independent study of 36 relapsing–remitting multiple sclerosis patients. The median central : peripheral Gd-enhancing $T_1$ lesion volume ratio was not significantly different ($P = 0.9$) for lesions detected with single-dose (ratio $= 0.67$) and triple-dose (ratio $= 0.76$) contrast.

Longitudinal $T_1$ Gd-enhancing lesion probability maps from weekly scans

Another potential explanation for the observed distribution asymmetry in $T_2$ and Gd-enhancing LPMs is that periventricular Gd enhancement may simply be shorter-lived than that in the more peripheral white matter. With infrequent scanning this could lead to the appearance of lower Gd-enhancing lesion volumes in the periventricular region. To test this possibility we generated $T_1$ Gd-enhancing LPMs using data from weekly scans performed on five multiple sclerosis patients over 3 months. There were 59 new Gd-enhancing $T_1$ lesions. The median central : peripheral lesion volume ratio again was not significantly different from unity (0.49, range 0.09–1.64). Only one patient had a greater volume of Gd-enhancing $T_1$ lesions in the periventricular compared with the peripheral white matter region.

Discussion

We used LPMs to define the spatial distribution of $T_1$ Gd-enhancing and $T_2$ lesion volumes in a population of patients...
with relapsing multiple sclerosis. This approach revealed a marked distributional asymmetry between central and peripheral white matter regions for T1 Gd-enhancing and T2 lesion volumes. We found a substantially higher probability density for T2 lesions in the central relative to the peripheral regions; this was not seen for T1 Gd-enhancing lesions, which were evenly distributed through the cerebral white matter.

To define better the basis of this phenomenon we generated longitudinal LPMs that define new T1 Gd-enhancing and new T2 lesion volume distributions. Quantitative analysis of longitudinal LPMs showed the same central : peripheral T1 Gd-enhancing/T2 lesion distribution asymmetry that was seen in the cross-sectional study. This suggests that the distribution asymmetry is not due to a consistent central-to-peripheral progression of new disease with time.

The spatial asymmetry between T2 and T1 Gd-enhancing lesions might arise from several possible mechanisms. We favour the hypothesis that periventricular T2 lesion volumes can increase de novo without previous local breakdown of the blood–brain barrier. Visual inspection of the new T1 Gd-enhancing and new T2 lesion LPMs gave the impression that non-enhancing new T2 lesions often reflected increases in confluent periventricular T2 lesion volumes rather than the formation of new discrete T2 lesions (data not shown). This type of lesion growth in the periventricular white matter could occur by mechanisms different from those responsible for discrete peripheral lesions, e.g. progressive gliosis with Wallerian degeneration of axons traversing more peripheral lesions. However, because the absolute number of T1 Gd-enhancing lesions is defined by the frequency of scanning and dose of contrast used (in contrast to their spatial distribution), determining with certainty on an individual basis precisely which lesions arose independently from Gd enhancement was not possible.

Alternative hypotheses for the asymmetry of T1/T2 lesion distribution include the possibility that the central white matter might have a greater susceptibility to persistent T2 hyperintense changes following inflammation. However, we did not find an increased likelihood of T1 Gd-enhancing lesions progressing to T2 lesions in the central compared with the peripheral white matter region. This is suggestive, though not conclusive, that preferential white matter susceptibility is insufficient to explain the spatial T2 lesion asymmetry found. Another possibility is that new T2 lesions in the periventricular region are less likely to resolve than are more peripheral lesions. We found no evidence for increased absolute volumes of T2 lesion resolution within the peripheral compared with the periventricular white matter region based on yearly T2 scans, although the data suggest that the amount of resolved T2 lesion as a proportion of new T2 lesion volume was somewhat less within the periventricular white matter region. A final possibility might be that Gd enhancement in the periventricular region could be more subtle or less long-lived than more peripheral enhancement, but the similarity in the distribution of T1 Gd enhancement from the principle study and from the triple-dose or weekly T1 Gd-enhanced scanning studies makes this unlikely.

Previous studies are not entirely inconsistent with the possibility that some T2 lesions may arise without prior T1 Gd enhancement. Bastianello and colleagues (Bastianello et al., 1990) scanned four relapsing patients monthly for 4 months and saw enhancement in 10 of 13 manually identified new T2 lesions. Thompson and colleagues (Thompson et al., 1992) scanned five patients with relapsing multiple sclerosis once every 2 weeks for 6 months and reported enhancement in only 77% of new T2 lesions. Later studies using larger patient groups and including both enlarging and new T2 lesion areas have also demonstrated that a significant proportion of T2-weighted signal change did not appear to be associated with prior Gd enhancement. In a two-centre study, Miller and colleagues (Miller et al., 1993) found that 15% of 144 new or enlarging T2 lesions in 26 patients did not show corresponding T1 Gd enhancement. Recent pathological studies also have suggested that some acute lesions may not be associated with breakdown of the blood–brain barrier (Gay et al., 1997). In patients with benign multiple sclerosis, Gd enhancement was seen in only 33% of all new T2 lesions (Thompson et al., 1992). The relatively high proportion of T2 lesion volume that appears to arise without prior Gd enhancement in our study may be a consequence of the increased sensitivity of methods employing accurate image registration for detection of subtle extension of the large, confluent periventricular lesions. This may specifically increase sensitivity to non-discrete T2 lesion volume changes.

There is compelling histopathological evidence that areas of Gd enhancement are associated with the presence of acute inflammation and demyelination, both in humans (Adams et al., 1985; Katz et al., 1990) and in animals (Kuharik et al., 1988; Hawkins et al., 1990). However, T2 hyperintense signal may also arise non-specifically from the progressive gliosis expected secondarily to Wallerian degeneration of axons. A notable anatomical feature of the periventricular white matter is the relatively high density of long projection tracts.

Corroborative evidence for possible heterogeneity in the mechanisms of T2 lesion genesis in multiple sclerosis comes indirectly from recent magnetic resonance spectroscopic imaging studies (Narayana et al., 1998) showing significant areas of lipid resonance in regions with no associated T1 Gd enhancement. These areas subsequently went on to develop new T2 lesions on longitudinal follow-up. Such findings suggest that demyelination may occur independently of perivenous inflammatory activity and support the possibility of more than one pathological process leading to tissue damage in multiple sclerosis. MRI studies in patients with different clinical forms of multiple sclerosis also highlight the possibility of heterogeneity in the genesis of T2 lesions.

The distributional asymmetry for T1 Gd-enhancing and T2 lesions was observed in our study using conventional T2 imaging parameters with a slice thickness of 5 mm. Recent studies have shown increased sensitivity for T2 lesion detection using thinner slices (Filippi et al., 1995b) and
FLAIR (fluid-attenuated inversion recovery) (Filippi et al., 1996; Yousry et al., 1997). If the more subtle T2 lesions detected with these newer techniques have a significant peripheral white matter preponderance, the degree of distribution asymmetry suggested by our findings might be an overestimate. However, it has not been reported that such methods detect substantially more peripheral than central white matter T2 lesions. FLAIR detects a greater number of subcortical T2 lesions, but also detects increased periventricular T2 lesion volumes. The poor sensitivity of FLAIR for infratentorial T2 lesions (Bastianello et al., 1997) may increase any central/peripheral white matter T2 lesion asymmetry. We were also careful to exclude cortical lesions from the analysis to reduce any mismatch between detection of cortical T1 Gd enhancement and cortical T2 lesion asymmetry. We were unable to detect any significant difference in the spatial distribution of T1 Gd-enhancing lesions detected with either triple-dose contrast or weekly scanning protocols compared with the more conventional protocol used in our multiple sclerosis study population. This suggests that, whilst the absolute number of T1 Gd-enhanced lesions is determined by scanning frequency and dose of contrast agent used, the spatial distribution of lesions detected remains essentially unchanged, i.e. the distribution of blood–brain barrier breakdown is approximately random within the CNS white matter.

Several issues need to be addressed in further development of LPMs for investigating the dynamics of MRI markers of pathology within the CNS. Good registration accuracy with the standard brain is important in making meaningful spatial comparisons between patients. In this study, accuracy of registration was assessed qualitatively by using software that allowed the observer to toggle between the registered image and the standard brain to ensure overlap of landmark anatomy. Subtraction images were also generated for the standardized image volumes to create null images as a further check for areas of poor tissue registration. Such visually qualitative techniques are reliable and have been validated elsewhere (Wong et al., 1997). In this study the spatial comparisons were of low resolution, concerning merely a large central and peripheral white matter region, and should therefore have been relatively insensitive to small errors in registration. Confidence in making higher-resolution spatial comparisons, such as for the assessment of relative lesion loads in the posterior limb of the internal capsule between different patient groups, would be enhanced by using isotropic voxel or three-dimensional acquisition MRI techniques that would allow higher precision registration with the standard brain. To generate the longitudinal new T2 lesion LPMs, accurate intra-patient registration was initially required to produce the difference images for each patient. Accurate intra-patient registration was aided by attention to precise inter-scan repositioning. Intra-patient registration is therefore required only to match morphologically similar images differing slightly in spatial orientation. As such, intra-patient registration errors were small and caused difference image lesion volume errors of <0.5%. This technique for producing accurate difference images has been described previously, and potentially offers a more sensitive method for monitoring changes in T2 lesion activity (Lee et al., 1998).

Understanding the heterogeneity of T2 lesion genesis is important if we are to interpret accurately MRI-monitored treatment trials, and may help us understand the discrepancy observed between clinical disability and T2 lesion load. The possibility that some T2 lesions may arise de novo also provides support for the concept that there are multiple mechanisms for lesion generation in multiple sclerosis. The coexistence of different mechanisms of disease progression would have important implications for both current and new therapeutic interventions. We believe that LPMs provide a simple yet powerful method for investigating the spatial and temporal dynamics of MRI markers relevant to the understanding of pathology in multiple sclerosis and potentially also that of other neurological conditions (Miyai et al., 1997).

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References
Filippi M, Paty DW, Kappos L, Barkhof F, Compston DA, Thompson AJ, et al. Correlations between changes in disability and T2-


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