**Magnetic Resonance Imaging of Animal Models of Encephalitis**

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The use of animal models has been fundamental to advancing our understanding of the neuroinflammatory processes associated with several neurological disorders, including stroke, multiple sclerosis, amyotrophic lateral sclerosis, Parkinson’s disease and Alzheimer’s disease. Multiple sclerosis (MS) is a relapsing remitting and/or chronic progressive inflammatory disease of the human central nervous system (CNS) that is characterized by inflammation, demyelination, axonal injury, and neurodegeneration. MS is the most common chronic inflammatory demyelinating disease affecting young adults in Western countries (1). Although the specific causes of MS are, to this date, unknown, it is well accepted that its progress involves autoimmune reactions against the main constituents of the CNS (2,3). The autoimmune aspects of MS are experimentally modeled in laboratory animals following the induction of autoimmunity to CNS antigens. The neurological disorder that develops is called experimental autoimmune encephalomyelitis (EAE). Rodent and non-human primate models have served as invaluable tools to investigate the mechanisms of the immune response against self-antigens within the CNS, as well as to test new therapies for the treatment of autoimmune diseases (4).

In particular, common marmosets (*Callithrix jacchus*), a New World primate model native to Brazil, have been extensively used as a model of human MS. Common marmosets are small (300-500 g body weight) animals that breed well in captivity and reach sexual maturity by 18 months of age, allowing for a relatively easy maintenance of in-house outbred colonies at substantially lower costs than other nonhuman primate species. In common marmosets, EAE may be induced through injection of whole myelin, myelin extracts in complete Freund’s adjuvant (CFA), and human recombinant myelin oligodendrocyte glycoprotein (rh-MOG) in CFA (5-7). Clinical signs of disease in marmosets more closely resemble MS than do those in rodent or other primate models and follow a relapsing-remitting/secondary progressive course. The progression of clinical signs may involve the typical asynchronous formation and development over time of lesions anywhere in the CNS, that is, lesions are disseminated in time and space (8). Thus, the same individual may present a variety of lesion stages. Furthermore, while neurological signs are usually associated with lesion onset, there is a noticeable discrepancy between the appearance of clinical symptoms and the lesion load detected with brain MRI, a phenomenon known as
Magnetic resonance imaging has been an excellent tool to visualize pathological changes in CNS tissues (10). In human patients with MS, MRI reveals macroscopic tissue abnormalities with high sensitivity. MS lesions appear hypointense in T1-weighted sequences such as MPRAGE (10). Lesions are isointense in proton-density (PD)-weighted MRI, but hyperintense in T2- and T2*-weighted MRI, or in T1-weighted MRI post gadolinium (Gd) administration. The most commonly used diagnostic MRI techniques have been implemented in the marmoset EAE model. In our laboratory, we use a protocol which includes MPRAGE, PD, T2W, T2*W, T1-mapping and T1W with pre- and post-gadolinium contrast. T2W and T2*W have a high sensitivity for detection of MS lesions, which appear as focal areas of hyperintensity, while the PD shows isointense lesions and the MPRAGE shows hypointense lesions (Fig. 1). The T2W images can be used to determine the total number and size of lesions. However, T2W images provide little information about to the underlying pathological status of the lesions. T2*W MRI is used to add information about the presence of vascular structures, in particular veins, around the lesions. It is know that MS lesions develop around small, inflamed veins (11). To detect acute lesions with active inflammation, it is best to use contrast-enhanced MRI. Gd-enhanced T1W MRI allow distinction of active lesions from inactive ones, as inflammation results in increased blood-brain barrier permeability. Leakage of Gd causes delineation of the active lesions. According to their pattern of contrast enhancement on static MRI, lesions can be classified as nodular or ringlike. Ringlike lesions are associated with more severe tissue damage due to demyelination and axonal loss. However, static MRI cannot capture the spatiotemporal expansion of lesions. Using dynamic contrast-enhanced MRI, Gaitan and colleagues recently reported that lesions with initial nodular enhancement in human MS patients are smaller than those with initial ringlike presentation (11). Furthermore, following Gd administration, all nodular lesions enhance centrifugally, whereas ringlike lesions enhance centripetally, becoming nodular or nearly nodular (11).

Fig. 1: Examples of MR images of a marmoset with EAE lesions. From left to right, lesions appear isointense in the Proton Density (PD) MRI, hyperintense in the T2- and T2*-weighted MRI, and hypointense in the MPRAGE MRI.
This rapid change of enhancement dynamics from centrifugal to centripetal reflects the outward growth of MS lesions around their central vein and suggests that factors mediating lesion growth and tissue repair derive from different locations at different times (11).

In the marmoset EAE model, using high magnetic field strengths (≥ 7T), MRI can be performed in vivo at high spatial resolution (~ 300 µm isotropic) within clinically acceptable times. At such resolution, white matter lesions can be detected several weeks before the onset of clinical symptoms. Ringlike lesions can be observed as well, confirming that the lesions form around small veins. In addition, quantitative T1 measurements before and after Gd-injection reveal increased BBB permeability within normally appearing white matter (NAWM) early during the course of the disease. Relatively large permeability changes precede the development of mostly small lesions. When scanning post-mortem excised brain and spinal cord specimens, the spatial resolution can be decreased even further (100 µm isotropic). Interestingly, microscopic lesions can be detected post-mortem that were not detected in equivalent scans in vivo at 300 µm, raising the question on whether permeability changes in NAWM are actually due to the early presence of such microscopic lesions.

In conclusion, MRI has been successfully used in the marmoset EAE model to address 2 important issues: 1) to evaluate disease activity in the CNS in presymptomatic animals; 2) to investigate the relationships between MRI changes and defined neuropathologic changes. It will be interesting to examine potential pathological differences between NAWM areas and lesion areas at such early timepoints, in the hope of better establishing the onset of inflammation prior to the detection of the lesions in vivo.

References