Treatment with a Human Antibody That Promotes Remyelination Results In Decreased Lesion Load as Detected by T2 Weighted MRI in a Viral Model of MS

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Purpose:

Multiple Sclerosis (MS) is a demyelinating disease of the central nervous system (CNS). The final outcome of a demyelinating lesion is either scar tissue formation or regenerative repair. Stimulation of remyelination may be a viable restorative treatment approach. Our laboratory has identified both human and mouse monoclonal antibodies which promote remyelination in animal models of MS.

MRI is a well-established tool in the diagnosing of human MS patients. The aim of our study was to determine whether changes in T2 weighted lesion load would allow us to characterize the antibody-mediated remyelination in SJL/J mice with TMEV- induced demyelinating disease. These animals develop a progressive demyelinating disease, with lesions mostly confined to the spinal cord.

Materials and methods

Theiler's Virus Infection of mice

TMEV induced demyelination was produced by intracerebral virus injection of 4-8 week old SJL/J mice. All animals develop mild viral encephalitis, which resolves within 14 days. The animals then develop a chronic demyelinating disease in the spinal cord that progresses over several months.

Treatment with remyelinating antibody

Thirteen animals 6 months post TMEV infection received a single intraperitoneal injection of 0.5 mg of the previously characterized sHIgM22 antibody in PBS, another eight received PBS alone.

Magnetic resonance imaging (MRI)

The mice were followed by serial MRI-s. Images were obtained on the day before treatment, and 5 weeks thereafter. Previous studies have demonstrated that remyelination will be almost complete by then. The MRI examinations were performed in a Bruker Avance 300 MHz (7 Tesla) vertical bore NMR spectrometer equipped with "miniimaging" accessories. We used a custom-manufactured mouse holder for stabilizing the animals. T2 weighted imaging was performed using a RARE volume acquisition sequence (TR: 2000ms, TE: 70ms, Flipback:on, RARE factor: 16, FOV: 3.5cm x 3.5 cm x 3.5 cm, matrix: 160x160x160, resolution: 218.75 µm isometric).

Analysis of 3D image sets: spinal cord extraction, co-registration, segmentation, and volumetry

Analyze 5.0 (Biomedical Imaging Resource, Mayo Clinic) was used for image analysis. The spinal cords were extracted using the Object Extractor and Image Edit tools. A 3D voxel-matching algorithms was used for co-registration. The extracted and co-registered spinal cord segments were then cropped to include the cervical cord and the first 4 thoracic segments only. The individual images were then segmented using the 3D ROI Analysis Tool. A semi-automated intensity-based seed-growing algorithm was used to generate object maps. These object maps defined numbered, individually identified, three-dimensionally represented cord lesions. The generated object maps were then applied to the image set representing the subsequent time point, and were corrected as appropriate with the same seed-growing algorithm so that changes in lesion number, volume and shape could be accounted for. To minimize the "human factor", the involved investigators were trained for several weeks on test datasets, and their intra-and inter-rater reliability was was found to be superior (>95%). After the generation of object maps, the 3D ROI Scan Tool was used to calculate lesion volumes.

Results:

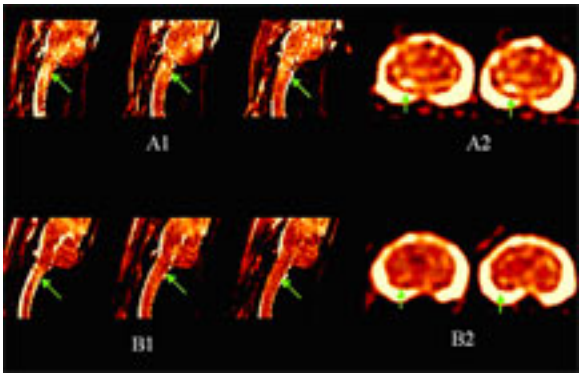
HslgM22 antibody reduced total T2 lesion load within five weeks of treatment.

Mean lesion loads of the treated and non-treated groups were not significantly different before treatment (406.5 (109.2) vs 460.9 (122.6), p=0.37). However, five weeks after treatment the mean lesion loads in the two groups became different (241.4 (85.5) vs 523.4 (112.7), p=0.0000027). The mean lesion load decreased by 40.6% in the treated group, whereas it increased by 13.6% in sham-treated animals. We found that 18.75% of lesions in the control group showed signs of retraction. In the antibody treated group, we found that 82.8% of lesions showed retraction of varying degrees.

Discussion:

Previous histopathology studies using this particular antibody demonstrated that remyelination occurred in 59.7% of spinal cord lesions compared to 15.8% in PBS treated animals. The reason for this could be the higher sensitivity of volumetric MRI compared to two-dimensional random-sampling histopathology methods.

Our results indicate that T2 weighted 3D volume-acquisition MRI was capable of capturing remyelination in this virus-induced model of MS. At present, there is no universally accepted marker for remyelination using standard MRI techniques. T2 weighted sequences are considered sensitive, but not specific for demyelinating disease. In some models, the resolution of T1 hypointensities correlated with remyelination, in others, increasing magnetization transfer ratio was found to correlate with newly forming myelin. In our model, T1 hypointensities were not observed. However, we did observe hypointense lesions on T2 weighted sequences, adjacent to the hyperintense lesions. These were especially prominent in the animals treated with the remyelinating antibody, thus their presence may be related to remyelination. The observed decrease in the T2 hyperintense lesion volume may be related to a "masking" effect caused by the adjacent T2 hypointensities. We conclude that the applied 3D RARE T2 sequence is a sensitive measure of remyelination in this animal model of MS.



Sagittal (A1, B1) and axial (A2, B2) spinal cord MRI image samplesof the same mouse. Image slices were reconstructed from the original 3D datasets, and pseudo-colored using "hotiron" encoding. A1, A2: spinal cord images before antibody treatment; B1, B2: spinal cord images 5 weeks after antibody treatment. The green arrows point to a large area of T2 hyperintense lesion in the upper cervical cord. The lesion size visibly decreases on the post-treatment images.