Modulated focused ultrasound for treatment of de-myelinating axons in multiple sclerosis lesions - pilot animal studies

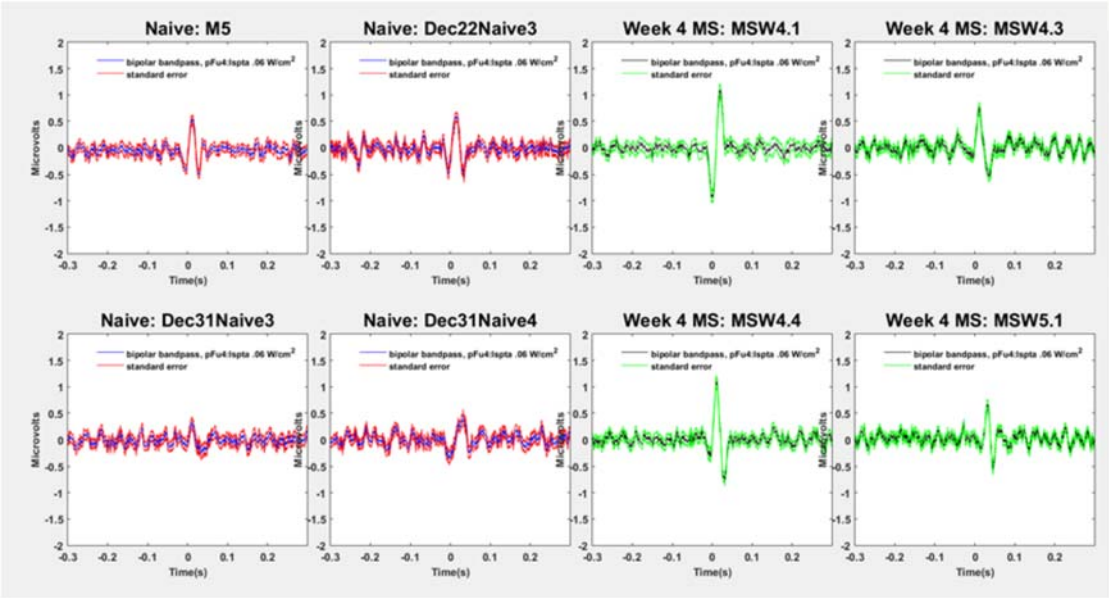
6 month report, June 1, 2015 – December 31, 2015

This project seeks to assess the ability of pulsed focused ultrasound (pFU) to slow down or stop de-myelination as well as accelerate re-myelination in a mouse model for multiple sclerosis (MS).

For Aim #1 we have identified two pre-existing focused ultrasound transducers with frequencies (2 MHz and 1.1 MHz) close or identical to those of existing MRI-guided HIFU systems. We started design work for a third transducer to run at a frequency of 0.65 MHz. We have characterized the 2 MHz device in water and identified a number of ultrasound protocols to apply to MS mouse brains. The 1.1 and 0.65 MHz transducers will be characterized in this way as well.

Achieving IACUC approval for the study took some time owing to its complicated nature. Full IACUC approval was achieved in August 2015. A researcher was trained on the use of the MRI in August 2015 and performed initial MRI trials in October and November 2015.

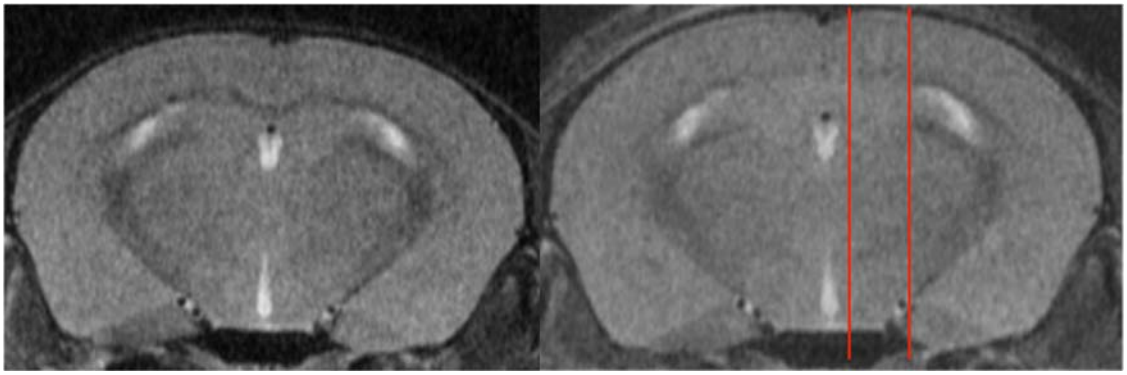
We established the cuprizone MS model. Specifically, de-myelination occurs in mice after chronic consumption of a copper-based food known as cuprizone. During September-November 2015 we tested various protocols with the 2 MHz device on normal (non-MS) mice and MS mice undergoing de-myelination, described below. With that work we identified an optimal ultrasound protocol: one that generated robust and reliable neural activation as verified by EEG and fell below FDA limits in terms of intensity. Additionally, we compared the EEG activity generated by the same protocols in normal mice and MS mice (figure one, overleaf). We found that the shape of pFU-induced activity peaks generated in each group of mice differ from each other, though we observed that a given ultrasound protocol always worked in each of normal and MS mice. Specifically, with the same ultrasound we generated smaller EEG signals in normal mice (red figures below) relative to MS mice (green figures below).



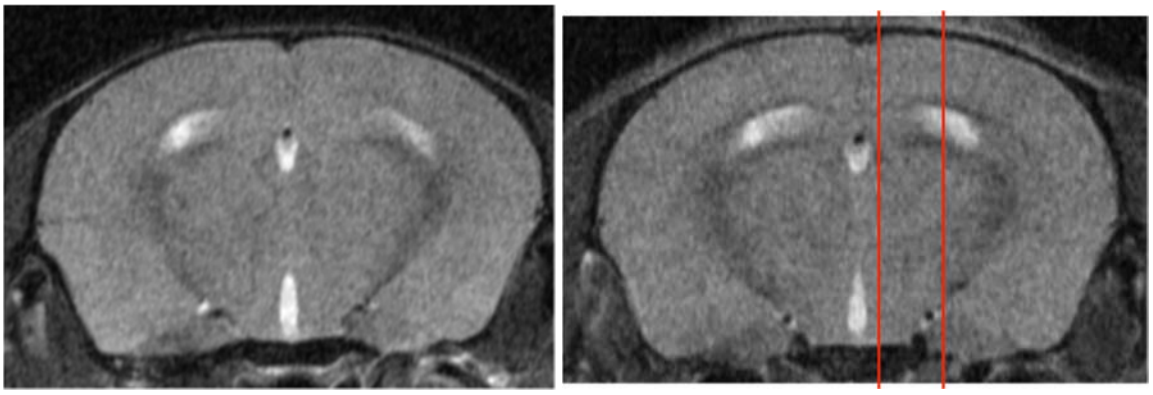
Average and variance of EEG signals [vertical axis] collected from four normal

mice (red) and four MS mice (green) through time [horizontal axis, spanning from -0.3 seconds to 0.3 seconds], the latter undergoing chemically induced de-myelination. Ultrasound applied at time = zero generates a spike in electrical activity starting at the time of ultrasound application.

During the first six months of our work we established 2 groups of mice, one in October 2015 (the long term group, during which the corpus callosum re-myelinates after ten weeks of de-myelination) and one in November 2015 (the shorter term group, during which the corpus callosum de-myelinates over a three week period). During week four we applied a pFU protocol daily for 5 days using the 2 MHz device to mice undergoing the short-term MS model prior (Aim #2). The mice continued on cuprizone throughout the week of ultrasound application and for one week after. Below (left), MRI image of the brain of a short-term MS model mouse before ultrasound application. Below (right), the same brain after ultrasound application, which was focused between the two red lines.



We applied a pFU protocol daily for 5 days using the 2 MHz device to mice undergoing the MS model for 10 weeks and 2 weeks of recovery, prior to pFU application (Aim #3). Before and after the week of ultrasound application, MRI scans were performed. Below (left), the brain of a 10-week MS model mouse before ultrasound application. Below (right), the same brain after ultrasound application, which was focused between the two red lines.



In the second half of this yearlong project we will apply pFU protocols using the 0.65 and 1.1 MHz ultrasound devices to the brains of mice undergoing the 3 and 10 week MS models. We will also complete histological analysis of all brains that received a therapeutic pFU protocol, using a myelin stain to assess the effects of ultrasound on re-myelination. Finally, we have added quantitative MRI imaging protocols to our study, to increase the signal-to-noise level in the corpus callosum.