***Assessment of splenic perfusion and blood flow of SMA model mice***

*Background and goal*

We have recently uncovered major abnormalities in the spleens in our spinal muscular atrophy model mice. Many potential defects might be simultaneously occurring, one of which we hypothesized to be abnormal blood perfusion of the spleen.

Therefore, the goal of this study is to investigate blood flow to and in the spleen by doppler ultrasound using the VEVO2100.

*Methods*

Wild-type and SMA model mice will be subjected to doppler Ultrasound of the spleen at constant interval (at least 7 days apart) to better understand vasculature development and progression. The *Smn2B/-* model mice have onset of motor phenotype at around P12 and a median lifespan of 25 days. Spleen defects are first apparent at P4 and become progressively worse with time. Therefore, we aim to perform 3 ultrasounds maximum on the course of their lifespan (more specifically at P4, P11 and P19) for both wild-type and *Smn2B/-* model mice.

During this procedure, the mouse will be anesthetized using 2% isofluorane. Tear gel will be applied to avoid drying of the mouse’s eyes. It will then be placed on the amovible stage, where it will be shaved (from mid stomach to hips) with a clipper and further application of Nair to ensure complete hair removal. The mouse will then be restrained lightly by applying a small piece of tape on each paw in order for them to touch an electrode for cardiac monitoring. This will ensure optimized anesthetic condition for the mice to be comfortable. Respiratory rate will also be monitored for the same purpose. Procedure may be variable in time depending on the ease to identify splenic artery and vein in the animal. Once appropriate measurements are taken, the mice will be returned to their cage for recovery.

We do not anticipate any adverse events on the mice from this procedure.

\*Hair removal may also be done prior to the experiment day to optimize ultrasound machine usage.