Final Progress Report – Lay Summary **March 2019**

Development and Validation of an MS-MS Method for Detection of Hexosaminidase Deficiency in Tay Sachs Disease

Our laboratory has developed two novel methods for measuring hexosaminidase enzyme activity, the enzyme deficient in Tay Sachs (TS) and Sandhoff (SD) diseases. We are also in the process of developing a third method which uses completely automated sample processing and analysis. This last method will be the most suitable for mass screening of newborns once a treatment becomes available and screening is approved for TS and SD.

We started developing a method by tandem mass spectrometry for measuring TS and SD using a synthetic substrate from a Dr Gelb is Seattle. It turned out that this was not suitable for the purpose that we intended it. As a result, we switched to using MUG and MUGS, two compounds that have been used in the past to measure hexosaminidase enzyme activity. The hexosaminidase enzyme breaks down MUG or MUGS to produce 4-MU, a chemical that can be measured easily by a variety of methods. All of our methods will therefore measure 4-MU, the product of the reaction between MUG or MUGS and the hexosaminidase enzyme to detect TS and or SD. All three methods we have developed and validated (or still developing) using either mass spectrometric, or fluorescence detection of 4MU.

The completely automated assay we are still working on is in collaboration with a Dr Henderson at Children's Hospital of Eastern Ontario, the home of the Ontario Newborn Screening Laboratory.

A second aim of the proposed project was to collect 5000 newborn screening blood spot samples from an area of the Province of Quebec that has a high incidence of Tay Sachs disease. We will determine the carrier frequency of TS disease in this population by measuring hexosaminidase enzyme activity, and also confirm the presence or absence of the mutated gene in these samples by a method called Polymerase Chain Reaction (PCR).

We have so far only received two fifth of the 5000 samples we are going to analyze from Quebec, but expect the remaining samples to be shipped to us in the next few days or weeks. These will be analyzed as soon as they are received.

To date we have analyzed ~2000 samples sent to us from Quebec. Our results show that blood from newborn babies is stable once it is collected on filter paper, and can be stored and analyzed even 3-4 years after blood collection. Hexosaminidase enzyme activity is stable if the samples are stored in the freezer, but slowly decline if stored at room temperature. The mutations and the methods for measuring them in the Quebec population of TS patients have been published. These same methods will be used by our laboratory. We have measured Hexosaminidase enzyme activity in 2000 samples collected between 2015-2016. Enzyme activity was normally distributed, and averaged between 6-700 nanomoles /liter/hour, the commonly used unit to express enzyme activity.

In conclusion, our laboratory has developed methods suitable for mass screening of hexosaminidase activity in dried blood spots, the sample normally used for newborn screening. Once all the samples have been analyzed, we will publish our results in appropriate journals, which will make these methods available for other labs to set up these methods for mass screening of TS and SD.

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