January, 2012

Six Months Progress Report: Studies of Taurine-conjugated GM2 in Tay-Sachs Disease

By Yu-Teh Li, Ph.D., Professor, Department of Biochemistry and Molecular Biology , Tulane University School of Medicine

1) Development of an enzyme-linked immunosorbent assay (ELISA) for the detection of taurine-conjugated GM2 (TGM2)

Q: What is the reason for studying taurine-conjugated GM2 (TGM2) in Tay-Sachs brain?

A: i) It is well known that one of the hallmarks of Tay-Sachs disease is the massive accumulation of GM2 (a sugar linked lipid) in the central nervous system (CNS). The obvious question is:
How does the CNS react to and cope with the massive GM2 accumulation? To answer this question, in 2003 we methodically studied gangliosides (a group of sugar linked lipids) in normal and Tay-Sachs brain samples and detected the presence of TGM2 (taurine attached to GM2) in Tay-Sachs brains, but not in normal brains.

ii) Taurine is a unique and very water-soluble amino acid widely found in animal tissues. Since taurine conjugation is a well known detoxification mechanism in biological systems to facilitate the clearance of xenobiotics (chemical compounds that are foreign to a living organism) from the body by increasing their water solubility, we hypothesized that Tay-Sachs brains may regard the massively accumulated GM2 as a foreign material and employ taurine conjugation as a vehicle for its removal.

iii) To understand the pathobiological significance of TGM2, such as the biosynthesis and removal of this unusual GM2 derivative from the CNS (slide 1), it is essential to have a sensitive method for the detection of TGM2 and we chose to develop an ELISA for this purpose. ELISA is a very sensitive analytical method based on antigen-antibody interaction. The principle of ELISA was first described in 1960 by Rosalyn Yalow (a 1977 Nobel laureate) for the detection of plasma insulin concentration.
For ELISA analysis of TGM2, we have collaborated with the University of California Davis/NIH Neuro Monoclonal Antibody Facility (through the support of the NIH) to successfully generate an anti-TGM2 monoclonal antibody (mAb). Using this mAb and our chemically synthesized TGM2, we have established the basic parameters for the analysis of TGM2 by ELISA during the past six months.

2) Analysis of GM2 accumulation at different locations of a Tay-Sachs brain

Q: What is the merit for analyzing GM2 accumulation at different locations of a Tay-Sachs brain?

A: Due to the limited availability of autopsy brain samples from Tay-Sachs patients, the traditional analysis of GM2 accumulation in a Tay-Sachs brain was performed by using a small piece of brain sample taken from an autopsy brain without considering the original location of the sample in the brain. Based on immunostaining, Phaneuf *et al.* (Human Molecular Genetics, 5, 1-14, 1996), found that the distributions of GM2 in Sandhoff mouse brain varied widely from region to region.
We reasoned that the accumulation of GM2 in human Tay-Sachs brains might also be "location- specific". Since we have a whole freeze-dried brain in slices from a juvenile variant B Tay-Sachs patient (stored in a -80 °C freezer), we carried out the analysis of the distribution of GM2 at twenty different locations of this brain. We found that the extent of GM2 accumulation differed considerably among the twenty different locations (slide 2). The accumulation of GM2 was found to be most profound at the four locations taken from cerebellum (slide 3). We feel this is a very important observation, as TGM2 in Tay-Sachs brains could also be associated with the regional distribution of GM2. The reason for the variation in GM2 accumulation at different locations of a Tay-Sachs brain remains to be established.