## 00310 Splice-Switching Oligonucleotides (SSOs) Specific for Modulating Fyn Isoforms: A New Tool for Alzheimer's Disease Research

*Lim Ting Wei Frances, Lee Ching Li, Low Ying Bei Clara, Tan Guet Khim Michelle*

Singapore General Hospital

**Aims:** Fyn tyrosine kinase is a potential therapeutic target for Alzheimer's disease (AD). Our lab demonstrated that upregulation of FynT in neurofibrillary tangle bearing neurons was concomitantly found with decreased FynB, suggesting that dysregulation of mutually exclusive splicing machinery may induce splicing switch from FynB to FynT in AD brain. The aim of the study is to assess whether Splice-Switching Oligonucleotides (SSOs) can be used for manipulating alternative splicing of Fyn to achieve desirable outcomes.

**Methodology:** Exonic splicing enhancers and silencers were first identified by Human Splicing Finder software as the target sequences for the design of 3 independent FynB-SSOs and 4 independent FynT-SSOs. SSOs were first tested for cytotoxicity and their ability to induce alternative splice switching in transfected cells. Selected SSOs were then introduced to primary rat neurons to monitor the isoform specific role of Fyn in response to Amyloid-ß (25-35) treatment.

**Result:** The SSOs were low in cytotoxicity and able to induce alternative splice switching effectively, as shown by FynB-SSOs induced FynB-specific exon skipping to favour FynT expression and FynT-SSOs induced FynT-specific exon skipping to favour FynB expression. Stronger immunoreactivity of autophosphorylated Fyn was detected in FynB-SSO transfected cells, suggesting that FynT has stronger kinase activity. Primary rat neurons transfected with FynB-SSOs presented a higher FynT expression were more susceptible to Amyloid-ß (25-35) treatment.

**Conclusion:** FynB-SSOs and FynT-SSOs have been proven to induce splice switching of Fyn isoforms effectively. FynB-SSOs can be used to generate neurons with predominant FynT expression, for better understanding the signalling pathways in associated with the progression of AD. On the other hand, FynT-SSOs can be served as a potential therapeutic treatment, to specific target the pathogenic FynT in AD.