The Role of IA-2 Extracellular Domain in Type 1 Diabetes Progression

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Purpose: The tyrosine phosphatase-like protein (IA-2) is a major autoantigen associated with the progression and detection of T1D. We identified a new candidate biomarker within the extracellular domain of IA-2 (IA-2ec) in humans, which leads to rapid acceleration of T1D onset compared to conventional IA-2 biomarkers of T1D. Our objective is to identify autoantibody binding domains specific for IA-2ec, in order to determine their pathophysiological impact on cell function. Our central hypothesis is that autoantibodies present during the progression of T1D are pathogenic and cause distinct physiological alterations in cellular metabolism.

Methods: We synthesized and assayed a new bio-marker construct against the human IA-2ec, amino acids 26-577, through standard serum autoantobody screening via radio-immunoassay (RIA). Competitive bindings studies utilizing IA-2ec and standard bio-markers associated with the progression of T1D were also carried out to fully characterize IA-2ec recognition. Human SH-SY5Y neuroblastoma cells were differentiated and treated with poly-clonal (Santa Cruz) IA-2ec antibodies, wherein they were assayed to determine possible physiological effects on calcium channel activity and calcium mobilization upon antibody treatment. Lastly, nPOD subjects previously identified as reactive for conventional IA-2 or IA-2 derived bio-markers were screened for the presence of IA-2ec via RIA.

Summary of Results: We developed a new bio-marker construct against the human IA-2ec, which successfully identifies IA-2ec positive individuals through standard serum RIA. This response is associated with a high risk of progression toward T1D. Competitive binding experiments further support the presence of autoantibody responses directed toward the extracellular domain of IA-2. Upon treatment with poly-clonal IA-2ec antibodies, differentiated SH-SY5Y cells displayed significant increases in mobilized calcium and calcium channel activity compared to isotype treated controls. In addition, a small set of nPOD subjects previously identified as being positive for conventional IA-2 based biomarkers, were able to be identified as positive for IA-2ec autoantiboides.

Conclusions: Our data shows that the extracellualr domain of IA-2 is a novel antigenic determinant associated with rapid progression to T1D. Furthermore, antibodies reactive toward IA-2ec have the ability to cause distinct physiological alterations in calcium channel signaling and the mobilization of calcium. Future studies utilizing IA-2ec postive nPOD subjects will be carried out to elucidate IA-2ec autoantibody binding sequences to afford the production of monoclonal antibodies and Fab fragments specific for IA-2ec recognition.