From Genetics to Gene Expression in nPOD Tissues

Disease-dependent and Tissue-specific Changes in Gene Expression in Type 1 Diabetes (T1D)

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Purpose: Use microarray global gene expression profiling to compare mRNA expression in different tissues (pancreatic lymph node, spleen) from T1D patients and normal subjects to identify genes whose expression is disease-dependent and tissue-specific.

Methods: To identify differentially expressed genes in T1D verses healthy controls, we performed cDNA microarray analysis on pancreatic lymph node (pLN) and splenic tissues from 10 T1D patients and 10 healthy controls. These tissues were snap frozen in RNAlater and total RNA was extracted using standard Trizol methods. All frozen tissue samples were kindly provided by nPOD. Gene expression was analyzed using an Agilent 41K Whole Human Genome (60-mer) Oligo Microarry Kit. Statistical and pathway analyses of differentially modulated genes in T1D were performed using Genespring and Ingenuity Pathway Analysis (IPA) software tools, respectively.

Summary of Results: Large-scale gene expression analysis of T1D patients' tissues compared to healthy controls revealed disease-dependent and tissue-specific modulation of genes. Despite the heterogeneity in global gene expression in individual samples, disease-dependent 1532 entities with over annotated 1000 genes were specifically modulated by at least 1.5X in pLN of T1D patients while only 459 entities or slightly over 300 genes were changed in the spleen. These disease-dependent modulated genes were also tissue-specific since there is little overlap between genes differentially expressed in the pLN and spleen. Most differentially expressed genes in the spleen are non-immune specific while those in the pLN are enriched for a network of immune- and inflammation-related genes that clearly distinguish T1D from healthy individuals. These data question the notion raised by SNP and GWAS studies proposing that specific fixed mutations in disease-relevant genes are a major cause of T1D. Rather than global genetic defects in all tissues and cells as SNPs and GWAS mutations would propose, our data support the concept that disease-specific changes in gene expression occur in specific tissues such as the pLN. In particular, the genes expressed differentially in the pLN are involved in regulatory pathways including MHC molecules, NFkB, TCR, apoptosis, PI3K and Stat proteins whose expression is altered in T1D compared to controls. Remarkably, a significant number of these genes are seen in in cytotoxic T cell mediated apoptosis, antigen presentation, OX40 signaling and cell cycle regulation.

Conclusions: Global gene expression analysis of T1D patients versus healthy controls revealed disease-dependent and tissue-specific gene signatures. Key changes in the expression of genes relevant to immune and inflammation-specific pathways were seen in the pLN. Overall, tissue specific gene expression in T1D was primarily associated with cell cycle, immune regulation, transcriptional regulation and signal transduction. These novel gene signatures and their role in immune regulation may provide insights into the pathogenesis of T1D and possibly provide targets for immunotherapy.