During development, GDNF signalling through the Ret receptor tyrosine kinase is critical for the initial evagination of the ureteric bud (UB) from the Wolffian duct for its subsequent branching and morphogenesis to give rise to the collecting system of the kidney. Downstream of the GDNF/Ret signalling cascade, the two ETS transcription factors Etv4 and Etv5 are activated in the tip cells of the UB. Both Etv4 and Etv5 are jointly required for kidney development. However, the mechanisms by which Etv4 and Etv5 regulate the cellular responses that lead to UB branching remain to be fully elucidated. By carrying out microarray screens comparing Etv4−/−;Etv5−/− mutant kidneys with wild-type kidneys, several putative target genes of these transcription factors were identified. Many of these genes, including Slc04c1, Vsnl1, Krt23, Lmnc2 and the metalloproteinase pair Adamts18 and Adamts16, showed UB-specific or UB-tip specific expression in wild-type kidneys and were either absent or down-regulated in the mutant kidneys. Furthermore, many of these genes have evolutionarily conserved ETS-binding sites in their promoter and enhancer elements, enabling chromatin immunoprecipitation studies to identify whether these candidate genes are direct targets of Etv4 and/or Etv5. Overall identification of novel genes downstream of Etv4 and Etv5 with previously unknown roles in kidney organogenesis could potentially help explain certain human renal birth defects at a molecular/genetic level.

**Program/Abstract # 208**

**Role of Etv4 and Etv5 in pancreatic development**

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FGF10 is a key signaling factor involved in the mesenchymal-epithelial interactions of many developing organs and tissues. In the embryonic pancreas, FGF10 is secreted from the mesenchyme to expand the progenitor population in the neighboring pancreatic epithelium. While Fgf10 is crucial for active proliferation of the pancreatic progenitors, few of its downstream genes have been identified. In an Fgf10 overexpression model, we found that FGF10 upregulated expression of two novel ETS-family transcription factors, Etv4 and Etv5, in the pancreatic progenitors. In wild type embryos, both factors are expressed almost exclusively within the pancreatic epithelial progenitors. We examined the pancreas of Etv4 and Etv5 null embryos to ascertain which effects of Fgf10 are mediated through these two factors. Etv5 null embryos showed a reduction in total pancreatic mass. The mature cell types were also diminished in the Etv5 null embryos. The pancreas of Etv4 null embryos appeared normal. However, genomic-profiling of the Etv4 null pancreas revealed reduced expression of factors necessary for endocrine and exocrine cell development, although this effect was not as severe as observed in the Etv5 null embryos. Considering that the DNA binding domains of these factors are almost identical, we conclude that genetic redundancy between these factors likely exists. This is currently being tested using compound breeding of Etv4 and Etv5 knockout mice.

**Program/Abstract # 209**

**Hox6 genes are important niche factors that play critical roles in the proper formation and maintenance of the pancreas**

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Despite significant advances in our understanding of endocrine pancreatic development, the function of the pancreatic mesodermal niche in this process is less well understood. However, niche factors are necessary for proper endocrine development and are likely to be of critical importance in designing successful regenerative therapies aimed at replacing lost islet cells in diabetic patients. Preliminary data generated in our laboratory demonstrates a critical role for Hox6 genes in pancreatic organogenesis. Hox6 genes are expressed exclusively in the pancreatic mesoderm (and not endoderm) and suggest a primary role for Hox6 genes in proper development of the pancreatic niche. The pancreatic phenotypic abnormalities observed in our Hox6 triple mutants confirm this, as total pancreatic volume in mutants is reduced compared to littermate controls and there is a greater than 90% reduction in insulin-expressing cells. In addition, insulin- and glucagon-expressing cells do not form proper islets and are abnormally positioned within the pancreas. Finally, while triple mutants die shortly after birth, surviving compound mutants exhibit hyperglycemia and impaired responses in glucose tolerance tests. Moreover, these defects are exacerbated with age and, as Hox6 genes remain expressed in the pancreas through post-natal and adult stages, suggest that Hox6 genes may contribute to post-natal endocrine maintenance as well. Overall, these data suggest that Hox6 genes are critical pancreatic niche factors, necessary for the proper development and maintenance of pancreatic organogenesis.

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**Program/Abstract # 210**

**Notch mediated patterning and cell fate allocation of pancreatic progenitor cells**

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Early pancreatic morphogenesis is characterized by the transformation of an uncommitted pool of pancreatic progenitor cells into a branched pancreatic epithelium that consists of “tip” and “trunk” domains. These domains have distinct molecular signatures and differentiate into different pancreatic cell lineages. Cells at the branched tips of the epithelium develop into acinar cells, while cells in the trunk subcompartment differentiate into endocrine and duct cells. Recent genetic analyses have highlighted the role of key transcriptional regulators in the specification of these subcompartments. Here, we analyzed the role of Notch signaling in patterning of the pancreatic epithelium through mosaic overexpression of a Notch signaling antagonist, dominant negative mastermind-like (dnMAML1), resulting in a mixture of wild type and Notch-suppressed pancreatic progenitor cells. Relative to the wild type cells, the Notch suppressed cells lose “trunk” maker genes and gain expression of “tip” genes. These cells undergo a process of sorting and rearrangement, leading to positioning of the Notch suppressed cells at the tip of the branched pancreatic epithelium, while the wild type cells occupy the “trunk”. The Notch suppressed cells subsequently differentiate into acinar cells, while duct and endocrine are formed predominantly from the wild type cells.