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The *Irx3* and *Irx5* genes in inner ear development

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Iroquois genes encode a family of highly conserved homeodomain transcription factors across the metazoans that are involved in multiple developing processes. *Irx3* and *Irx5* are linked in IrxB cluster in mouse and human genome, which are essential in the neurogenesis of ventral neural tube. Preliminary data indicated that *Irx3* and *Irx5* double mutant mice display significant hearing defect. However, the role of these two genes during inner ear development is poorly characterized. To understand the function of *Irx3* and *Irx5* during mouse inner ear development, the expression of these two genes in the otic placode and derivatives are examined. Two allelic mouse mutants, *Irx3tauLacZ* and *Irx3flox;Irx5EGFP,* which labeled *Irx3* and *Irx5* by β-gal and EGFP respectively, are analyzed to investigate the expression of these two genes. *Irx3* is expressed in the ventral region of otic vesicle at E10.5 and *Irx5* is expressed in the prosensory region of the cochlear duct at E12.5. Further experiments are currently conducted to complete the analysis of expression patterns of *Irx3* and *Irx5* during inner ear development.
Previous studies have demonstrated that *Irx3* and *Irx5* shares strikingly similar expression pattern, which could be explained by enhancer sharing and coregulation. To test this hypothesis, we attempt to identify the cis-regulatory elements of *Irx3* and *Irx5* which are responsible for inner ear development. Four putative inner ear specific enhancer elements are predicted in the intergenic region between *Irx3* and *Irx5* (~550kb) by phylogenetic anaylsis and transcription factor binding sites prediction. To examine the enhancer activities of these four regions, we have generated construct c1 to c4 with EGFP as a reporter gene and the chick in ovo electroporation assay will be performed.