EXAMINATION OF GENE MARKERS OF URETHRAL DEVELOPMENT IN SONIC HEDGEHOG SIGNALING DEFICIENT MICE

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INTRODUCTION AND OBJECTIVE: Incontinence is a common problem, and it imposes significant healthcare costs. The genetic basis of incontinence is poorly understood. Sonic hedgehog (Shh) signaling is a key signaling pathway in lower urinary tract development. Mice with reduced sonic hedgehog signaling display an incontinent phenotype. Our objective is to test the hypothesis that mice with reduced Shh signaling fail to form a distinct boundary between the bladder and urethra and therefore have inappropriately expressed gene markers in the epithelium of the bladder and urethra.

METHODS: Gli mutant mice (Gli2+/-; Gli3Δ699/+), were used as a Shh deficient mouse model. Female Gli mutant mice display an incontinent phenotype with GU malformations. Adult Gli mutant female mice and wild type controls were sacrificed and the lower urinary tracts, including the bladder and urethra, were dissected and embedded in paraffin. We compared bladder and urethra histology in Gli mutant and wild type mice. We immunostained Gli mutant and wild type female mouse bladders and urethras for cytokeratin 13 (KRT13) and transformation related protein 63 (TRP63) and compared the frequencies of stained cells.

RESULTS: There were no significant differences in the percentages of epithelial cells positive for TRP63, KRT13 or both markers between adult Gli mutants and wild-type controls. However, trending differences were observed. The mutant bladders showed higher percentages of TRP63 positive epithelium than controls. The mutant urethras showed lower percentages of TRP63 positive epithelium, higher percentages of double positive epithelium in the bladder neck region, and lower percentages of double positive epithelium in distal urethra than controls. Increasing the sample size and examining early developmental timepoints for these same epithelial markers is ongoing.

CONCLUSIONS: Our data indicate that there are trends toward differences in the differentiation of the epithelium of the urethra and bladder between Gli mutant mice and wild-type controls. This will indicate that mutations leading to reduced Shh signaling result in abnormal differentiation of the urethra epithelium which could lead to reduced ability for the urethra to function leading to incontinent phenotypes observed in these mice. People that suffer from genetically based incontinence or genital malformations often have Gli mutations. Better understanding the impact of Gli mutations on urinary tract physiology can lead to better treatment outcomes.

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