Downregulation of O-GlcNAc Levels Reduces Cilia Lengths and Renal Cystogenesis in ADPKD

Matthew A Kavanaugh, BS;[1, 4] Wei Wang, PhD;[1, 4] Henry H Wang, BS;[1, 4] Brittany M Jack, BS;[1, 4] Chad Slawson, PhD; [2, 4] Stephen C Parnell, PhD; [2, 4] Darren P Wallace, PhD; [3, 4] Pamela V Tran, PhD;[1, 4]
[1]Department of Anatomy and Cell Biology; [2]Department of Biochemistry and Molecular Biology; [3]Department of Molecular and Integrative Physiology; [4]Jared Grantham Kidney Institute, University of Kansas Medical Center, Kansas City, KS

Introduction: Primary cilia and cellular metabolism are important modifiers of ADPKD, and studies suggest their functions connect. Renal cysts occur in both ciliopathies and mitochondrial diseases. Additionally, renal primary cilia have been demonstrated to transduce fluid flow into metabolic pathways. The nutrient sensor, O-linked β-N-acetylglucosamine (O-GlcNAc), regulates mitochondrial function as well as ciliary homeostasis. Thus, we hypothesized that O-GlcNAc signaling is misregulated in PKD.

Methods: Immunostaining and Western blots were performed to examine O-GlcNAc levels in mouse and patient ADPKD tissue. Using the HoxB7-Cre in mice, *Pkd1* was deleted in collecting ducts, alone and together with O-GlcNAc transferase (*Ogt)*, which transfers O-GlcNAc onto protein substrates, and kidneys were analyzed on postnatal day 21. ADPKD patient cells were treated with pharmacological inhibitors of OGT and ciliogenesis to examine cilia lengths and *in vitro* cyst formation. Co-immunoprecipitation in *Pkd1* conditional knock-out (cko) renal tissue was performed to determine the endogenous targets of OGT.

Results: O-GlcNAc levels were elevated in mouse and patient ADPKD renal cyst-lining cells. *Ogt* deletion in juvenile *Pkd1* cko mice reduced ciliary lengths, renal cystogenesis, kidney weight:body weight ratios and improved kidney function. OGT inhibition in cultured ADPKD patient renal epithelial cells also shortened primary cilia and reduced cyst formation. Further, combined treatment of ADPKD cells with OGT and ciliogenesis inhibitors reduced cyst formation to a greater extent than treatment with either inhibitor alone. Preliminary co-immunoprecipitation experiments in mouse kidney lysates reveal OGT interacts with IFT81 and acetylated α-tubulin, and these interactions were increased in *Pkd1* cko kidneys.

Conclusions: O-GlcNAc is elevated in ADPKD and its downregulation reduces ciliary lengths and renal cystogenesis. OGT may target specific ciliary proteins in PKD, promoting increased cilia lengths. We propose that O-GlcNAc links the ciliary and metabolic defects in ADPKD and may present new avenues for designing therapeutic strategies.

Funding Sources: P30GM12273