O-GlcNAcase Inhibition Slows PKD Development

Chad Slawson*#, Zhen Zhang*, Miranda Machacek*, Ee Phie Tan*, Gentry Cork*, John Kelsh*, Stephen Jackson#, and Stephen Parnell*#.

Department of Biochemistry and Molecular Biology* and Jared Grantham Kidney Institute#, University of Kansas Medical Center.

INTRODUCTION

Although PKD is caused by inherited mutations; a full understanding of the cellular effects of the disease is poorly understood slowing drug development. We contend that O-GlcNAcylation is involved in PKD progression and that pharmacological interventions targeting O-GlcNAcylation could lead to novel therapies for PKD. O- GlcNAcylation is a ubiquitous post-translational modification of a single N- acetylglucosamine sugar attached to serine or threonine residues in nuclear, mitochondrial, and cytoplasmic proteins. The modification is rapidly processed to modulate O-GlcNAc homeostasis in response to changes in the cellular environment by O-GlcNAc transferase (OGT), which adds the modification, and O-GlcNAcase (OGA), which removes the modification.

METHODS

Using animal models of cystic kidney disease, we obtained control or cystic kidneys from a conditional mouse model at 7 or 14 days post-birth (P) and measured O- GlcNAcylation and OGT/OGA proteins by western blot or mRNA levels by qPCR. Additionally, we treated control and slowly-progressive $Pkd1^{RC/V}$ animals with 50 mg/kg of OGA inhibitor Thiamet-G (TMG) intraparietal injected every other morning for 15 days for a total of 8 doses per mouse, collected kidneys as before, and performed western blots or histological staining.

RESULTS

By P14, OGT expression and O-GlcNAc levels were lower and OGA levels were higher in conditional cystic animals compared to wild-type control animals. OGT mRNA drastically declined in the cystic compared to wild-type animals at P14. OGA mRNA from cystic animals was similar to control except for P7 in which OGA mRNA levels were lower in the cystic animals. Cystic $Pkd1^{RC/V}$ animals treated with TMG had half the KW/BW (kidney weight to body weight) ratio average compared to vehicle treated animals. No differences were observed in the control animals treated with TMG. As expected, TMG treated cystic kidneys showed less cystic area than the non-treated kidneys. Control and TMG-treated control kidneys had no morphological differences.

CONCLUSIONS

These data demonstrate that as cystic disease progresses O-GlcNAc levels and OGT protein and mRNA expression declines while OGA expression increases suggesting that as cysts develop, normal cellular O-GlcNAc homeostasis is disrupted Importantly, OGA inhibition reduced cystic growth demonstrating the potential for modulating O-GlcNAc homeostasis to slow PKD progression.