A Novel Porcine Model of ARPKD for Preclinical Studies Produced by Gene Editing.

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INTRODUCTION

Autosomal recessive polycystic kidney disease (ARPKD) is the most common genetic pediatric cystic renal disease affecting approximately 1 in 20,000 individuals. ARPKD is caused by a broad range of biallelic mutations in the *PKHD1* gene, including splice site, missense, and truncation mutations. Patients present with varying degrees of renal enlargement, renal cysts, hepatic fibrosis, and portal hypertension. Neonatal ARPKD mortality is 30%, and many patients succumb to end stage renal or hepatic disease before reaching adulthood. Mouse *Pkhd1* models develop very mild kidney disease and the rat ARPKD model is phenotypically similar to ADPKD, so no ideal model for ARPKD is available. The similarities between pigs and humans in anatomy, physiology, genetics, and size, specifically regarding cardiovascular and urological systems, make them an ideal animal to closely recapitulate human ARPKD for therapeutic development.

METHODS

To develop a swine model of ARPKD recapitulating the spectrum of human renal and hepatic disease severity, we performed cytoplasmic zygote injections of gene-editing reagents targeting *PKHD1* exon 3 and transferred them to recipient gilts to produce swine with various combinations of ARPKD alleles including the orthologous human T36M allele, frame-shift mutations, and in-frame indels in the same litter. These animals are being characterized by monthly blood chemistry, urine chemistry, and ultrasound or MRI. When disease severity meets our criteria for euthanasia or by 6 months of age, animals will be euthanized and samples collected for histological, gene expression, and proteomic analysis.

RESULTS

Twenty-two piglets are being characterized with a range of phenotypes and genotypes from homozygous inactivating to single in-frame variants. Initial histology of early disease-presenting animals bears a striking resemblance to ARPKD patient histology. Animals with heterozygous mutations have been identified as breeders that could propagate the lines for future preclinical studies.

CONCLUSIONS

Our approach allowed the production of pigs with a spectrum of ARPKD severity from a single litter, with disease phenotypes closely resembling the human disease. This approach could be useful for producing ARPKD animals for preclinical studies, as well as to produce founder animals for a breeding population to study specific ARPKD mutations.