

**TITLE:** Cleavage of Pkhd1 results in a small fragment that localizes and functions within mitochondria

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**Introduction:** Autosomal recessive polycystic kidney disease is caused by mutations in PKHD1 encoding FPC, and is characterized by severe renal cystogenesis in neonates, yet mouse models do not fully recapitulate the human phenotype.

**Methods:** We use mouse models, biochemistry, cell models, and electron microscopy to reveal novel cleavage fragments of FPC and their effect on enhancing cystogenesis in Pkd1 mutant cyst-sensitized mice.

**Results:** We describe how Pkhd1 mutation modifies a Pkd1 uncleavable mutant (Pkd1V), enhancing the cystic phenotype in kidney and pancreas. FPC displays differential cleavage to produce fragments of unknown function. We identify several of these cleavage fragments and describe the mitochondrial localization of one small C-terminal fragment, induced by a newly identified mitochondria localizing signal presented in the fragments. We found mitochondrial ultrastructural changes after deletion of Pkhd1 including mitochondrial fragmentation and dilated cristae, indicating disrupted mitochondrial function. Finally, we show that deletion of just the C-terminal fragment of FPC ( $\Delta$ CT) is sufficient to enhance the renal cystic phenotype of the PC1 cleavage mutant but does not result in the pancreatic cystogenesis seen in other Pkhd1 mutants on this background.

**Conclusion:** Our results suggest that the C-terminus of FPC plays an important role in preventing cystogenesis via a novel mitochondria specific function.

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