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Polycystic Kidney Disease: New Knowledge and Future Promises

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Abstract

Polycystic kidney disease (PKD) is one of the most common genetic kidney diseases, characterized by the formation of fluid-filled renal cysts, which eventually leads to end-stage renal disease. Despite several decades of investigation, the explicit molecular and cellular mechanisms underpinning renal cyst formation have been until recently unresolved, severely hampering the development of effective therapeutic approaches. Currently, most PKD therapies have been developed for limiting disease complications, such as hypertension. Although Tolvaptan has been approved for treating PKD in few countries, the associated hepatic toxicity remains a major concern. In this Review, we will discuss the most recent advances in PKD research, covering aspects ranging from newly identified genetic/epigenetic causes, increment in mechanistic interpretation, novel therapeutic targets, to the promises offered by the emerging stem cell technology.

Introduction

Polycystic kidney disease (PKD) is constituted by three major forms: autosomal dominant (ADPKD), autosomal recessive (ARPKD), and syndromic (such as Joubert, Meckel and Bardet Biedle syndromes) PKD, in which renal and liver cyst formation accounts for most of the morbidity [1]. ADPKD is usually adult-onset with an estimated occurrence of 1:500-1:1,000. Clinical studies estimate that mutations in *PKD1* (encoding Polycystin1) and *PKD2* (encoding Polycystin2) account for approximately 78% and 13% of ADPKD cases respectively, with the remaining 9% cases genetically unsolved [2]. ARPKD, mostly caused by *PHKD1* mutations, has an occurrence of 1:20,000, and often manifests *in utero* or in the perinatal period [2].

PKD has long been categorized as a ciliopathy, because most PKD-related loci are targeted to ciliary-centrosome complex. There has been a growing consensus on the possible factors contributing to cyst growth (Figure 1). Polycystin1 (PC1) and Polycystin2 (PC2) appear to constitute the foundation of cystic pathway. Genetic mutations within the central PKD machinery produce functionally defective proteins, leading to decreased intracellular Ca^{2+} and increased cyclic adenosine monophosphate (cAMP) levels, the two cellular hallmarks of PKD [2]. These early events remodel multiple pathways, including mTOR, LKB1-AMPK, JAK-STAT, Ras-MAPK, EGFR, PKA and WNT, the combinatory effects of which result in aberrant cell proliferation/apoptosis, increased trans-epithelial fluid secretion, faulty extracellular matrix (ECM) turnover, and metabolic reprogramming [3,4]. The recent advances in understanding the basis of PKD have both substantiated and challenged the existing theory of cyst formation. Here, we discuss these latest PKD studies, and the future directions that may lead to better understanding and novel therapeutics for the treatment of PKD.

New genetic/epigenetic causes

Whole exome sequencing (WES) has greatly facilitated the identification of novel genes associated with PKD. These novel genes encode proteins that interact with PC1 and PC2 in some way, and are important for maturation, trafficking, localization, and function of PKD proteins. These studies reiterated the importance of the central PKD machinery and highlighted novel regulators of Polycystin, which could serve as potential therapeutic targets. As one of the examples, *PKHD1* has long been considered as the only ARPKD gene until a recent landmark study described mutations in *DZIP1L* that encodes DAZ interacting protein 1-like in ARPKD patients [5*]. As a centriole protein, *DZIP1L* interacts with Septin2 to maintain the periciliary diffusion barrier at the ciliary transition zone [5*]. Loss of *DZIP1L* leads to bulged cilia, reduced ciliogenesis, as well as compromised ciliary translocation of PC1 and PC2 [5*,6]. Readers interested in other ciliary genes are referred to several excellent reviews on this topic [1,7-9].

It is noteworthy that several newly identified PKD genes are associated with endoplasmic reticulum (ER)-mediated protein homeostasis. One such gene is *GANAB*, which encodes the glucosidase II α subunit (GII α), a resident ER enzyme involved in asparagine (N)-linked glycosylation, which may participate in maturation and quality control of transmembrane glycoproteins. *GANAB* mutations were identified in patients that present with ADPKD or the related autosomal-dominant polycystic liver disease (ADPLD) [10-12]. *GANAB*^{-/-} cells showed defects in maturation, surface and ciliary localization of PC1 and PC2 [10]. Interestingly, in *GANAB*^{-/-} cells, either introduction of wild-type GII α or targeting of defective polyglutamylation restored PC2 localization, thus proposing a possible role of *GANAB* in cystogenesis [13]. WES also identified pathogenic mutations of *DNAJB11* in ADPKD. *DNAJB11*, a co-factor of BiP co-chaperone, has extensive functional overlap with *GANAB* in the maintenance of ER protein homeostasis. Thus, loss of *DNAJB11* results in defective maturation and trafficking of PC1 and UMOD [14*]. In addition, Cabezas *et al.* showed that a promoter mutation (c.-167G>T) in *PMM2*, which encodes, Phosphomannomutase 2, a key enzyme in N-glycosylation, may contribute to renal cyst formation through alteration of tissue-specific chromatin loop formation and transcriptional activity. While coding mutations in *PMM2* cause a congenital disorder of glycosylation with severe neurological symptoms, patients with this promoter mutation present with hyperinsulinemic hypoglycemia and congenital PKD (HIPKD) [15].

Epigenetic regulation represents another layer of regulation on the dose of gene products. Genome-wide methylation analysis described that hypermethylation of the functional copy of *PKD1* in ADPKD patients resulted in haploinsufficiency of *PKD1* gene products [16]. In ADPKD patients, significant epigenetic silencing was also seen in genes implicated in cellular transport, adhesion, and cell differentiation pathways [16]. A recent study demonstrated global hypomethylation in ADPKD cortical kidney tissues using single nucleotide resolution methylome profiling [17]. Increased methylation at the 3' end of *PKD1* gene body was also observed, although *PKD1* mRNA expression remained unchanged [17]. Single cell sequencing approaches may help to elucidate more subtle cell-level changes that correlate with disease onset and progression. Further evaluation of epigenetic and transcriptomic changes in PKD patients or disease models may reveal novel modifiers of cystogenesis that could serve as therapeutic

targets. For example, Li *et al.* identified lysine methyltransferase SMYD2 as an epigenetic modifier that facilitates PKD pathogenesis in *PKD1* knockout mice via modulation of the methylation status of p53, p65, STAT3, and histones [18].

Old pathways, new interpretation

New findings have both supported and challenged the cilia-centered hypothesis, in which primary cilia function as a mechanosensor to regulate intracellular Ca^{2+} concentration via PC1 and PC2 complex-mediated Ca^{2+} currents. Kim *et al.* provided supportive evidence that WNTs bind to the extracellular domain of PC1 and induce Ca^{2+} currents in a PC2-dependent manner [19]. However, work by the Clapham group showed that PKD1L1 and PKD2L1, instead of PC1 and PC2, form a heteromeric ion channel to control ciliary Ca^{2+} concentration without altering global cytoplasmic Ca^{2+} concentration, in a flow-insensitive manner [20,21]. Recently, the Clapham group demonstrated a complete lack of mechanically induced Ca^{2+} increase in primary cilia, further challenging the ciliary hypothesis [22]. Future study may need to consider combinatory modes of Ca^{2+} channel sensing and activation in addition to mechanosensation or ligand binding. Furthermore, it has been intensively discussed whether primary cilia safeguard against or facilitate cystogenesis. Disruption of ciliary intraflagellar transport (IFT) leads to renal cyst formation in non-orthologous mouse models [23]. On the contrary, ablation of primary cilia [24], or induction of cilia disassembly via Aurora-A activation [25], reduces cyst growth in *PKD1* and *PKD2* mutant mice.

It is not until recently had we started to be aware of the negative feedback loop between primary cilia and autophagy. Defective ciliary IFT compromises nutrient deprivation-induced autophagosome biogenesis [26,27], while autophagy negatively regulates ciliogenesis, possibly through the autophagic degradation of OFD1, which promotes ciliogenesis [26,27]. Since impaired autophagic flux was observed in kidneys of ADPKD patients [26], autophagy activation may offer a promising strategy for cyst management [28].

Meanwhile, recent studies are gradually elucidating the complex regulatory network underpinning the defective cell proliferation and apoptosis occurring in PKD [29-32]. It was also revealed that multiple adenylyl cyclases (ACs) are implicated in the upregulation of intracellular cAMP. Loss or inhibition of ACs, in most cases, effectively ameliorates renal cyst formation in orthologous PKD and PLD mouse models [33-36]. Furthermore, last three years have witnessed an outbreak on the revelation of PKD protein/protein complex structure [37,38,39*,40,41]. A comprehensive structural analysis will provide a perfect means to bridge the knowledge gap in between genetic mutations and functional defects, as well as disease presentation in PKD.

Novel therapeutic target --- metabolic reprogramming in PKD

Over the last few years, accumulating evidence indicates that ADPKD shares many similarities with solid tumors despite overarching differences in disease presentation [42]. Among these, metabolic reprogramming has emerged as a novel hallmark of PKD [43]. In cancer, glucose metabolism is shifted from TCA cycle to glycolysis to meet high energy demand. In the context of PKD, it is similarly important for the cells to produce sufficient energy to sustain two essential cellular cystic phenotypes, proliferation and trans-epithelial fluid transportation.

In 2013, Rowe and colleagues demonstrated for the first time that *PKD1* mutations lead to dependence on enhanced glycolysis in ADPKD, via simultaneous upregulation of mTORC1 and inhibition of LKB1-AMPK signaling [44,45]. A follow up study further illustrates that PKD progression can be ameliorated upon chronic treatment of low doses of 2DG, which restores the levels of phosphorylated AMPK without obvious toxicity in the brain and heart [46]. Likewise, food restriction, an alternative approach to modulate energy metabolism, ameliorates PKD-related disease phenotypes, similarly through activating LKB1-AMPK as well as inhibiting mTOR pathway, without affecting the cellular levels of cAMP and SIRTUIN [46,47].

Consistent with increased glycolysis, reduction in lipid metabolism and oxidative phosphorylation (OXPHOS) was observed in PKD. *PKD1*^{-/-} mice display intrinsic dysfunction in fatty acid oxidation (FAO) [48]. Decreasing dietary lipid intake improves some of PKD-related phenotypes in *PKD1*^{-/-} mice [48]. Hajarnis *et al.* reported that in renal cysts of mouse and human ADPKD, reduction of OXPHOS and FAO may be attributed to miR-17-mediated suppression of *Pparα* [49*]. Anti-miR-17 treatment attenuates cyst growth in two PKD mouse models (*PKD1*^{RC/RC}, and *Nphp3*^{pcy/pcy}), as well as an *in vitro* human cell culture model of PKD, thus offering a promising therapeutic approach [49*].

Aberrant glutamine metabolism has recently been described in both murine ARPKD and ADPKD models. It is plausible that a glutamine-dependent TCA shunt towards 2-hydroxyglutarate (2-HG) production serves to fuel cell growth and proliferation during cystogenesis [50]. Flowers and colleagues further demonstrated that both *LKB1* and *PKD1* mutant mice display glutamine-dependent ureteric growth and branching morphogenesis, which can be rescued by adding either non-essential amino acids or by reducing glutathione [51*]. Inhibition of glutamine metabolism greatly retards cyst progression in both orthologous and non-orthologous PKD mouse models *in vivo* [51*,52*].

There have been accumulating efforts to address the correlation between metabolic reprogramming with the PKD machinery. There is evidence that lack of PC1 expression may mimic a low oxygen environment, leading to decreased ER-mediated Ca²⁺ release [53]. Subsequently, defective Ca²⁺ signaling results in oxidative stress through reduced PGC-1α (peroxisome proliferator-activated receptor γ coactivator 1α) expression and increased cAMP-PKA signaling [54]. Clinical studies have identified oxidative stress early in ADPKD, which can be reduced by administering antioxidants [54]. Several recent studies proposed that Bicc1 (Bicaudal C1), a non-classic PKD protein, is implicated in multiple metabolic pathways, including mTOR and gluconeogenesis [29,55]. It is conceivable that metabolic rewiring may be a common phenotype shared by various types of cystic kidney diseases. We are only just starting to shed new lights on the roles of metabolic reprogramming in PKD (Figure 2). It will be exciting if we could re-purpose drugs that are currently used to target cancer or metabolic diseases for the treatment of PKD [56].

Stem cell-based PKD model

Over the last few years, tremendous efforts have been made to coax human pluripotent stem cells (PSCs), including both embryonic stem cells (ESCs) and induced PSCs (iPSCs), into three-dimensional (3D) organoids, which hold great promise in regenerative medicine [57]. Earlier studies successfully differentiated human PSCs into kidney progenitors that are capable of further maturing into 3D kidney structures upon organ co-culture [58,59]. More recently, several groups have successfully established protocols for differentiating human PSCs into complex 3D kidney organoids that are comprised of nephron-like structures patterned along a proximal-to-distal axis [60-64].

This approach offers unprecedented opportunities for studying human kidney development, disease pathogenesis, and to test drug toxicity and efficacy in a patient-specific manner (Figure 3). Morizani *et al.* provided proof-of-concept that in human PSC-derived kidney organoids, renal toxins damage specific nephron segments that are known to be susceptible to the tested drugs [61]. Meanwhile, the possibility of growing kidney organoids in a high-throughput manner greatly increases the feasibility and amenability in performing drug screening [65]. ADPKD patient iPSCs have been employed to study the cellular behavior of PC1 and PC2 upon differentiation into hepatic cells [66]. Freedman and colleagues have taken one step further by using CRISPR-Cas9 gene editing tools to generate *PKD1*^{-/-} and *PKD2*^{-/-} human PSCs that, upon differentiation into kidney organoids, form cysts spontaneously [67]. A recent study by the Freedman group has demonstrated remarkable cyst formation upon interference with the microenvironment of kidney organoids [68*]. The removal of adherent cues expedites cystogenesis in *PKD1*^{-/-} and *PKD2*^{-/-} kidney organoids [68*]. It is beyond any doubt that human PSC-derived kidney organoid platform will soon be widely utilized to model various aspects of human PKD pathogenesis.

Future perspectives

The progress made during the last five years has been remarkable, offering new insights as well as raising questions on our current understanding of PKD. Technological advances always lead to knowledge explosion. Single cell sequencing approaches may be harnessed to decipher the genetic/epigenetic regulations underpinning the sporadic cyst origin of PKD. In the foreseeable future, structure elucidation of PKD proteins (wildtype and mutants) will sketch a map that accurately correlates genetic lesions with the respective functional defects.

The discovery of metabolic reprogramming as a novel hallmark and therapeutic target of PKD has been encouraging. Several clinical trials are already in the pipeline. In many diseases, metabolic reprogramming represents a secondary effect and as a result, it may not be sufficient to cure PKD by modulating metabolism alone. It will be critical to dissect the interactions between metabolic reprogramming and the core PKD machinery, including the PC1 and PC2 complex as well as the primary cilia.

The discrepancy of findings between different studies strongly indicates the importance of model selection and validation. To this point, patient iPSC-derived kidney organoids offer both promises and challenges in our efforts towards personalized medicine. The current major concern is that most human PSC derivatives are fetus-like and immature. Recent single cell studies have also revealed missing cell types, as well as inter-organoid variation [69,70]. There is an urgent need

to consistently generate kidney organoids that harbor the desired cellular components with correlative functionality. A cross-consultation between directional differentiation [71] and organ-on-a-chip technology [63,72] may bring the current kidney organoid platform to the next level.

Conflict of interest statement

The authors declare no conflict of interest.

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Figure legends

Figure 1 Mechanisms underpinning polycystic kidney disease pathogenesis.

Schematic drawing that summarizes our current understanding of PKD pathogenesis from genetic causes to dysregulated pathways and cellular phenotypic outputs. Light blue squares indicate recent discoveries and arrows indicate upregulation or downregulation of the indicated pathway.

Figure 2 Current schematic of metabolic reprogramming in PKD.

In PKD, an increase in glycolysis, fatty acid synthesis, and glutamine metabolism is accompanied with a decrease in oxidative phosphorylation and fatty acid oxidation.

Figure 3 Proposed workflow for using stem cell-based models to study PKD.

Skin biopsies derived from PKD patients or healthy individuals are reprogrammed into iPSCs. CRISPR-Cas9 gene editing tools are employed to either correct the defective gene, or to introduce a genetic lesion that is associated with PKD. Kidney organoids derived from patient-specific iPSCs will be utilized for studying disease mechanisms or drug screening.

References

1. Hildebrandt F, Benzing T, Katsanis N: **Ciliopathies**. *N Engl J Med* 2011, **364**:1533-1543.
2. Harris PC, Torres VE: **Genetic mechanisms and signaling pathways in autosomal dominant polycystic kidney disease**. *J Clin Invest* 2014, **124**:2315-2324.
3. Torres VE, Harris PC: **Mechanisms of Disease: autosomal dominant and recessive polycystic kidney diseases**. *Nat Clin Pract Nephrol* 2006, **2**:40-55; quiz 55.
4. Ong ACM: **Making sense of polycystic kidney disease**. *Lancet* 2017, **389**:1780-1782.
5. Lu H, Galeano MCR, Ott E, Kaeslin G, Kausalya PJ, Kramer C, Ortiz-Bruchle N, Hilger N, Metzis V, Hiersche M, et al.: **Mutations in DZIP1L, which encodes a ciliary-transition-zone protein, cause autosomal recessive polycystic kidney disease**. *Nat Genet* 2017, **49**:1025-1034.
- * The study identified the first novel ARPKD locus besides PKHD1, demonstrating that ARPKD is not a homogeneous disorder, and DZIP1L may regulate the ciliary membrane translocation of PC1 and PC2.
6. Wang C, Li J, Takemaru KI, Jiang X, Xu G, Wang B: **Centrosomal protein Dzip1l binds Cby, promotes ciliary bud formation, and acts redundantly with Bromi to regulate ciliogenesis in the mouse**. *Development* 2018, **145**.
7. Novarino G, Akizu N, Gleeson JG: **Modeling human disease in humans: the ciliopathies**. *Cell* 2011, **147**:70-79.
8. Waters AM, Beales PL: **Ciliopathies: an expanding disease spectrum**. *Pediatr Nephrol* 2011, **26**:1039-1056.
9. Lee JE, Gleeson JG: **A systems-biology approach to understanding the ciliopathy disorders**. *Gemone Med* 2011, **3**:59-68.
10. Porath B, Gainullin VG, Cornec-Le Gall E, Dillinger EK, Heyer CM, Hopp K, Edwards ME, Madsen CD, Mauritz SR, Banks CJ, et al.: **Mutations in GANAB, Encoding the Glucosidase IIalpha Subunit, Cause Autosomal-Dominant Polycystic Kidney and Liver Disease**. *Am J Hum Genet* 2016, **98**:1193-1207.
11. Besse W, Dong K, Choi J, Punia S, Fedeles SV, Choi M, Gallagher AR, Huang EB, Gulati A, Knight J, et al.: **Isolated polycystic liver disease genes define effectors of polycystin-1 function**. *J Clin Invest* 2017, **127**:3558.
12. Besse W, Choi J, Ahram D, Mane S, Sanna-Cherchi S, Torres V, Somlo S: **A noncoding variant in GANAB explains isolated polycystic liver disease (PCLD) in a large family**. *Hum Mutat* 2018, **39**:378-382.
13. He K, Ma X, Xu T, Li Y, Hodge A, Zhang Q, Torline J, Huang Y, Zhao J, Ling K, et al.: **Axoneme polyglutamylation regulated by Joubert syndrome protein ARL13B controls ciliary targeting of signaling molecules**. *Nat Commun* 2018, **9**:3310.
14. Cornec-Le Gall E, Olson RJ, Besse W, Heyer CM, Gainullin VG, Smith JM, Audrezet MP, Hopp K, Porath B, Shi B, et al.: **Monoallelic Mutations to DNAJB11 Cause Atypical Autosomal-Dominant Polycystic Kidney Disease**. *Am J Hum Genet* 2018, **102**:832-844.
- * This study identified DNAJB11 as a novel ADPKD gene, highlighting its role as a regulator of PC1 maturation and trafficking in ER.
15. Cabezas OR, Flanagan SE, Stanescu H, Garcia-Martinez E, Caswell R, Lango-Allen H, Anton-Gamero M, Argente J, Bussell AM, Brandli A, et al.: **Polycystic Kidney Disease with**

Hyperinsulinemic Hypoglycemia Caused by a Promoter Mutation in Phosphomannomutase 2. *J Am Soc Nephrol* 2017, **28**:2529-2539.

16. Woo YM, Bae JB, Oh YH, Lee YG, Lee MJ, Park EY, Choi JK, Lee S, Shin Y, Lyu J, et al.: **Genome-wide methylation profiling of ADPKD identified epigenetically regulated genes associated with renal cyst development.** *Hum Genet* 2014, **133**:281-297.
17. Bowden SA, Rodger EJ, Bates M, Chatterjee A, Eccles MR, Stayner C: **Genome-Scale Single Nucleotide Resolution Analysis of DNA Methylation in Human Autosomal Dominant Polycystic Kidney Disease.** *Am J Nephrol* 2018, **48**:415-424.
18. Li LX, Fan LX, Zhou JX, Grantham JJ, Calvet JP, Sage J, Li X: **Lysine methyltransferase SMYD2 promotes cyst growth in autosomal dominant polycystic kidney disease.** *J Clin Invest* 2017, **127**:2751-2764.
- * This study identified lysine methyltransferase SMYD2 as a novel epigenetic regulator of ADPKD.
19. Kim S, Nie H, Nesin V, Tran U, Outeda P, Bai CX, Keeling J, Maskey D, Watnick T, Wessely O, et al.: **The polycystin complex mediates Wnt/Ca(2+) signalling.** *Nat Cell Biol* 2016, **18**:752-764.
20. DeCaen PG, Delling M, Vien TN, Clapham DE: **Direct recording and molecular identification of the calcium channel of primary cilia.** *Nature* 2013, **504**:315-318.
21. Delling M, DeCaen PG, Doerner JF, Febvay S, Clapham DE: **Primary cilia are specialized calcium signalling organelles.** *Nature* 2013, **504**:311-314.
22. Delling M, Indzhukulian AA, Liu X, Li Y, Xie T, Corey DP, Clapham DE: **Primary cilia are not calcium-responsive mechanosensors.** *Nature* 2016, **531**:656-660.
23. Happe H, Peters DJ: **Translational research in ADPKD: lessons from animal models.** *Nat Rev Nephrol* 2014, **10**:587-601.
24. Ma M, Tian X, Igarashi P, Pazour GJ, Somlo S: **Loss of cilia suppresses cyst growth in genetic models of autosomal dominant polycystic kidney disease.** *Nat Genet* 2013, **45**:1004-1012.
- * This study described for the first time that loss of intact cilia suppresses cyst growth following inactivation of polycystins, indicating divergent roles of primary cilia in cystogenesis.
25. Nikonova AS, Deneka AY, Kiseleva AA, Korobeynikov V, Gaponova A, Serebriiskii IG, Kopp MC, Hensley HH, Seeger-Nukpezah TN, Somlo S, et al.: **Ganetespib limits ciliation and cystogenesis in autosomal-dominant polycystic kidney disease (ADPKD).** *FASEB J* 2018, **32**:2735-2746.
26. Pampliega O, Orhon I, Patel B, Sridhar S, Diaz-Carretero A, Beau I, Codogno P, Satir BH, Satir P, Cuervo AM: **Functional interaction between autophagy and ciliogenesis.** *Nature* 2013, **502**:194-200.
27. Tang Z, Lin MG, Stowe TR, Chen S, Zhu M, Stearns T, Franco B, Zhong Q: **Autophagy promotes primary ciliogenesis by removing OFD1 from centriolar satellites.** *Nature* 2013, **502**:254-257.
28. Zhu P, Sieben CJ, Xu X, Harris PC, Lin X: **Autophagy activators suppress cystogenesis in an autosomal dominant polycystic kidney disease model.** *Hum Mol Genet* 2017, **26**:158-172.
29. Gamberi C, Hipfner DR, Trudel M, Lubell WD: **Bicaudal C mutation causes myc and TOR pathway up-regulation and polycystic kidney disease-like phenotypes in Drosophila.** *PLoS Genet* 2017, **13**:e1006694.

30. Wang E, Chiou YY, Jeng WY, Lin HK, Lin HH, Chin HJ, Leo Wang CK, Yu SS, Tsai SC, Chiang CY, et al.: **Overexpression of exogenous kidney-specific Ngal attenuates progressive cyst development and prolongs lifespan in a murine model of polycystic kidney disease.** *Kidney Int* 2017, **91**:412-422.
31. Parrot C, Kurbegovic A, Yao G, Couillard M, Cote O, Trudel M: **C-MYC is a regulator of the PKD1 gene and PC1-induced pathogenesis.** *Hum Mol Genet* 2018.
32. Conduit SE, Hakim S, Feeney SJ, Ooms LM, Dyson JM, Abud HE, Mitchell CA: **beta-catenin ablation exacerbates polycystic kidney disease progression.** *Hum Mol Genet* 2019, **28**:230-244.
33. Rees S, Kittikulsuth W, Roos K, Strait KA, Van Hoek A, Kohan DE: **Adenylyl cyclase 6 deficiency ameliorates polycystic kidney disease.** *J Am Soc Nephrol* 2014, **25**:232-237.
34. Johanns M, Lai YC, Hsu MF, Jacobs R, Vertommen D, Van Sande J, Dumont JE, Woods A, Carling D, Hue L, et al.: **AMPK antagonizes hepatic glucagon-stimulated cyclic AMP signalling via phosphorylation-induced activation of cyclic nucleotide phosphodiesterase 4B.** *Nat Commun* 2016, **7**:10856.
35. Spirli C, Mariotti V, Villani A, Fabris L, Fiorotto R, Strazzabosco M: **Adenylyl cyclase 5 links changes in calcium homeostasis to cAMP-dependent cyst growth in polycystic liver disease.** *J Hepatol* 2017, **66**:571-580.
36. Wang Q, Cobo-Stark P, Patel V, Somlo S, Han PL, Igarashi P: **Adenylyl cyclase 5 deficiency reduces renal cyclic AMP and cyst growth in an orthologous mouse model of polycystic kidney disease.** *Kidney Int* 2018, **93**:403-415.
37. Shen PS, Yang X, DeCaen PG, Liu X, Bulkley D, Clapham DE, Cao E: **The Structure of the Polycystic Kidney Disease Channel PKD2 in Lipid Nanodiscs.** *Cell* 2016, **167**:763-773 e711.
38. Grieben M, Pike AC, Shintre CA, Venturi E, El-Ajouz S, Tessitore A, Shrestha L, Mukhopadhyay S, Mahajan P, Chalk R, et al.: **Structure of the polycystic kidney disease TRP channel Polycystin-2 (PC2).** *Nat Struct Mol Biol* 2017, **24**:114-122.
39. Su Q, Hu F, Ge X, Lei J, Yu S, Wang T, Zhou Q, Mei C, Shi Y: **Structure of the human PKD1-PKD2 complex.** *Science* 2018, **361**.
- * This is the first cryo-electron microscopy structure of the truncated forms of human PKD1/PKD2 hetero-oligomeric complex at a near-atomic resolution (3.6 Å).
40. Su Q, Hu F, Liu Y, Ge X, Mei C, Yu S, Shen A, Zhou Q, Yan C, Lei J, et al.: **Cryo-EM structure of the polycystic kidney disease-like channel PKD2L1.** *Nat Commun* 2018, **9**:1192.
41. Zheng W, Yang X, Hu R, Cai R, Hofmann L, Wang Z, Hu Q, Bulkey D, Yu Y, et al.: **Hydrophobic pore gates regulate ion permeation in polycystic kidney disease 2 and 2L1 channels.** *Nat Commun* 2018, **9**:2302.
42. Seeger-Nukpezah T, Geynisman DM, Nikonova AS, Benzing T, Golemis EA: **The hallmarks of cancer: relevance to the pathogenesis of polycystic kidney disease.** *Nat Rev Nephrol* 2015, **11**:515-534.
43. Padovano V, Podrini C, Boletta A, Caplan MJ: **Metabolism and mitochondria in polycystic kidney disease research and therapy.** *Nat Rev Nephrol* 2018, **14**:678-687.
44. Priolo C, Henske EP: **Metabolic reprogramming in polycystic kidney disease.** *Nat Med* 2013, **19**:407-409.

45. Rowe I, Chiaravalli M, Mannella V, Ulisse V, Quilici G, Pema M, Song XW, Xu H, Mari S, Qian F, et al.: **Defective glucose metabolism in polycystic kidney disease identifies a new therapeutic strategy.** *Nat Med* 2013, **19**:488-493.
46. Chiaravalli M, Rowe I, Mannella V, Quilici G, Canu T, Bianchi V, Gurgone A, Antunes S, D'Adamo P, Esposito A, et al.: **2-Deoxy-d-Glucose Ameliorates PKD Progression.** *J Am Soc Nephrol* 2016, **27**:1958-1969.
47. Zhou X, Fan LX, Sweeney WE, Jr., Denu JM, Avner ED, Li X: **Sirtuin 1 inhibition delays cyst formation in autosomal-dominant polycystic kidney disease.** *J Clin Invest* 2013, **123**:3084-3098.
48. Menezes LF, Lin CC, Zhou F, Germino GG: **Fatty Acid Oxidation is Impaired in An Orthologous Mouse Model of Autosomal Dominant Polycystic Kidney Disease.** *EBioMedicine* 2016, **5**:183-192.
49. Hajarnis S, Lakhia R, Yheskel M, Williams D, Sorourian M, Liu X, Aboudehen K, Zhang S, Kersjes K, Galasso R, et al.: **microRNA-17 family promotes polycystic kidney disease progression through modulation of mitochondrial metabolism.** *Nat Commun* 2017, **8**:14395.
- * This study provided the first evidence that miR-17 unregulation contributes to ADPKD through Ppar α -mediated modulation of mitochondrial metabolism.
50. Hwang VJ, Kim J, Rand A, Yang C, Sturdivant S, Hammock B, Bell PD, Guay-Woodford LM, Weiss RH: **The cpk model of recessive PKD shows glutamine dependence associated with the production of the oncometabolite 2-hydroxyglutarate.** *Am J Physiol Renal Physiol* 2015, **309**:F492-498.
51. Flowers EM, Sudderth J, Zacharias L, Mernaugh G, Zent R, DeBerardinis RJ, Carroll TJ: **Lkb1 deficiency confers glutamine dependency in polycystic kidney disease.** *Nat Commun* 2018, **9**:814.
- * This research showed that PKD has developed an enhanced dependence on glutamine metabolisms, proposing a potential therapeutic target.
52. Podrini C, Rowe I, Pagliarini R, Costa ASH, Chiaravalli M, Di Meo I, Kim H, Distefano G, Tiranti V, Qian F, et al.: **Dissection of metabolic reprogramming in polycystic kidney disease reveals coordinated rewiring of bioenergetic pathways.** *Commun Biol* 2018, **1**:194.
- * This study revealed extensive rewiring of metabolic pathways in PKD, including enhanced glycolysis, glutamine metabolism, fatty acid synthesis, as well as reduced fatty acid oxidation and oxidative phosphorylation.
53. Padovano V, Kuo IY, Stavola LK, Aerni HR, Flaherty BJ, Chapin HC, Ma M, Somlo S, Boletta A, Ehrlich BE, et al.: **The polycystins are modulated by cellular oxygen-sensing pathways and regulate mitochondrial function.** *Mol Biol Cell* 2017, **28**:261-269.
54. Ishimoto Y, Inagi R, Yoshihara D, Kugita M, Nagao S, Shimizu A, Takeda N, Wake M, Honda K, Zhou J, et al.: **Mitochondrial Abnormality Facilitates Cyst Formation in Autosomal Dominant Polycystic Kidney Disease.** *Mol Cell Biol* 2017.
55. Leal-Esteban LC, Rothe B, Fortier S, Isenschmid M, Constam DB: **Role of Bicaudal C1 in renal gluconeogenesis and its novel interaction with the CTLH complex.** *PLoS Genet* 2018, **14**:e1007487.
56. Seliger SL, Abebe KZ, Hallows KR, Miskulin DC, Perrone RD, Watnick T, Bae KT: **A Randomized Clinical Trial of Metformin to Treat Autosomal Dominant Polycystic Kidney Disease.** *Am J Nephrol* 2018, **47**:352-360.

57. Clevers H: **Modeling Development and Disease with Organoids**. *Cell* 2016, **165**:1586-1597.
58. Xia Y, Nivet E, Sancho-Martinez I, Gallegos T, Suzuki K, Okamura D, Wu MZ, Dubova I, Esteban CR, Montserrat N, et al.: **Directed differentiation of human pluripotent cells to ureteric bud kidney progenitor-like cells**. *Nat Cell Biol* 2013, **15**:1507-1515.
59. Taguchi A, Kaku Y, Ohmori T, Sharmin S, Ogawa M, Sasaki H, Nishinakamura R: **Redefining the in vivo origin of metanephric nephron progenitors enables generation of complex kidney structures from pluripotent stem cells**. *Cell Stem Cell* 2014, **14**:53-67.
60. Takasato M, Er PX, Becroft M, Vanslambrouck JM, Stanley EG, Elefanty AG, Little MH: **Directing human embryonic stem cell differentiation towards a renal lineage generates a self-organizing kidney**. *Nat Cell Biol* 2014, **16**:118-126.
61. Morizane R, Lam AQ, Freedman BS, Kishi S, Valerius MT, Bonventre JV: **Nephron organoids derived from human pluripotent stem cells model kidney development and injury**. *Nat Biotechnol* 2015, **33**:1193-1200.
62. Takasato M, Er PX, Chiu HS, Maier B, Baillie GJ, Ferguson C, Parton RG, Wolvetang EJ, Roost MS, Chuva de Sousa Lopes SM, et al.: **Kidney organoids from human iPS cells contain multiple lineages and model human nephrogenesis**. *Nature* 2015, **526**:564-568.
63. Homan KA, Gupta N, Kroll KT, Kolesky DB, Skylar-Scott M, Miyoshi T, Mau D, Valerius MT, Ferrante T, Bonventre JV, et al.: **Flow-enhanced vascularization and maturation of kidney organoids in vitro**. *Nat Methods* 2019, **16**:255-262.
64. Garreta E, Prado P, Tarantino C, Oria R, Fanlo L, Marti E, Zalvidea D, Trepas X, Roca-Cusachs P, Gavalda-Navarro A, et al.: **Fine tuning the extracellular environment accelerates the derivation of kidney organoids from human pluripotent stem cells**. *Nat Mater* 2019, **18**:397-405.
65. Czerniecki SM, Cruz NM, Harder JL, Menon R, Annis J, Otto EA, Gulieva RE, Islas LV, Kim YK, Tran LM, et al.: **High-Throughput Screening Enhances Kidney Organoid Differentiation from Human Pluripotent Stem Cells and Enables Automated Multidimensional Phenotyping**. *Cell Stem Cell* 2018, **22**:929-940 e924.
66. Freedman BS, Lam AQ, Sundsbak JL, Iatrino R, Su X, Koon SJ, Wu M, Daheron L, Harris PC, Zhou J, et al.: **Reduced ciliary polycystin-2 in induced pluripotent stem cells from polycystic kidney disease patients with PKD1 mutations**. *J Am Soc Nephrol* 2013, **24**:1571-1586.
67. Freedman BS, Brooks CR, Lam AQ, Fu H, Morizane R, Agrawal V, Saad AF, Li MK, Hughes MR, Werff RV, et al.: **Modelling kidney disease with CRISPR-mutant kidney organoids derived from human pluripotent epiblast spheroids**. *Nat Commun* 2015, **6**:8715.
68. Cruz NM, Song X, Czerniecki SM, Gulieva RE, Churchill AJ, Kim YK, Winston K, Tran LM, Diaz MA, Fu H, et al.: **Organoid cystogenesis reveals a critical role of microenvironment in human polycystic kidney disease**. *Nat Mater* 2017, **16**:1112-1119.
- * This study demonstrated for the first time that removal of adherent environmental cues expedites cystogenesis within kidney organoids derived from *PKD^{-/-}* human pluripotent stem cells.
69. Wu H, Uchimura K, Donnelly EL, Kirita Y, Morris SA, Humphreys BD: **Comparative Analysis and Refinement of Human PSC-Derived Kidney Organoid Differentiation with Single-Cell Transcriptomics**. *Cell Stem Cell* 2018, **23**:869-881 e868.

70. Phipson B, Er PX, Combes AN, Forbes TA, Howden SE, Zappia L, Yen HJ, Lawlor KT, Hale LJ, Sun J, et al.: **Evaluation of variability in human kidney organoids.** *Nat Methods* 2019, **16**:79-87.
71. Taguchi A, Nishinakamura R: **Higher-Order Kidney Organogenesis from Pluripotent Stem Cells.** *Cell Stem Cell* 2017, **21**:730-746 e736.
72. Ronaldson-Bouchard K, Vunjak-Novakovic G: **Organs-on-a-Chip: A Fast Track for Engineered Human Tissues in Drug Development.** *Cell Stem Cell* 2018, **22**:310-324.