

Review Article

Moftah Mohamed Rabie

Department of Pediatrics & Pediatric Nephrology Faculty of Medicine, Al Azhar University, Cairo, Egypt.

Ciliopathies and Renal Diseases

ABSTRACT

Renal ciliopathies are a group of disorders characterized by nephronophthisis, cystic kidneys, or renal cystic dysplasia whose underlying disease pathogenesis is related to abnormal structure or function of the primary cilia complex. The number of renal ciliopathies continues to expand as genomic and genetic approaches identify novel causes. This in turn provides new opportunities to explore disease mechanisms and therapeutic approaches to target cystic kidney disease and other associated phenotypes. In the field of recent advances in renal ciliopathies, the novel genes for nephronophthisis (NPHP), autosomal dominant polycystic kidney disease (ADPKD), autosomal recessive polycystic kidney disease (ARPKD), and Joubert syndrome (JS) will be highlighted in this article, to improve our insight about the fascinating spectrum of these diseases.

KEYWORDS

Ciliopathies, Joubert syndrome, Nephronophthisis, autosomal dominant polycystic kidney disease, autosomal recessive polycystic kidney disease.

Corresponding author: Moftah M. Rabeea, MD Department of Pediatrics.
Faculty of Medicine, Al-Azhar University, Egypt.
Email : moftahmohamed11@hotmail.com

geget: The Journal of the Egyptian Society of Pediatric Nephrology and Transplantation (ESPNT)

geget <https://geget.journals.ekb.eg/>

Published by ESPNT <http://espnt.net/>

Cohosted by Egyptian Knowledge Bank <https://www.ekb.eg>

INTRODUCTION

Cilia are microtubule-based structures that protrude from the surface of almost every cell type in the human body. Cilia can be motile and be part of multiciliated structures or can be immotile, located as a solitary projection on the cell. Examples of motile cilia include the multiciliated cells lining the respiratory tract and the fallopian tube which beat in organized waves to move mucus or debris and the ovum respectively [1].

Immotile cilia, known as primary cilia, act as a cellular antenna coordinating cell signaling, polarity and other sensory and functional roles. Within the kidney, renal tubular cells exhibit primary cilia which project into the lumen of the nephron. Structurally, primary cilia consist of a basal body, a transition zone, and a ciliary axoneme (known as ciliary apparatus), which is highly conserved throughout many species **Figure 1&2**.

Ciliopathies are a group of multisystem diseases caused by mutations in genes encoding proteins located to ciliary apparatus. Ciliopathies in which the mutations lead to renal disease are known as renal ciliopathies [2]. The range of phenotypes associated with renal ciliopathies includes NPHP, cystic kidney disease and renal cystic dysplasia, often with renal interstitial fibrosis. Importantly, in renal ciliopathies extra renal features are often part of the patient phenotype and relate to cilia dysfunction in other tissues, most frequently the retina and brain. There is a growing list of these multisystem ciliopathies, including Joubert Syndrome, Jeune syndrome, and Bardet-Biedl syndrome [2].

The purpose of this review is to highlight recent molecular, genetic, and mechanistic studies concerning the common & clinically important renal ciliopathies (NPHP, ADPKD, ARPKD & JS) as a mean of updating a working biomedical and clinical knowledge on this topic.

Recent gene discoveries in renal ciliopathies

Several novel genes have recently been identified to cause renal ciliopathy syndromes including atypical forms of ADPKD, ARPKD-like syndromes, NPHP, and multisystem ciliopathy disorders such as JS **Table 1**

These discoveries emphasize the value of molecular genetic studies that employ next-generation sequencing techniques such as whole exome sequencing (WES). Indeed, the application of WES is being applied to many different cohorts of kidney diseases, with fruitful results [3] and it is expected that approach to become the ‘norm’. Identifying novel genetic causes of renal ciliopathies allows the opportunity to explore genotype-phenotype associations, novel disease mechanisms, and the cilium's fundamental biology **Figure 3**.

Novel genes for ADPKD: GANAB and DNAJB11

ADPKD is a well-known and established condition leading to polycystic kidney disease. It accounts for 10% of end-stage renal disease (ESRD). Clinical cohorts have been phenotypically studied in detail [4]. Over 90% of patients with ADPKD will have mutations in the known genes PKD1 or PKD2, but recently the

number of genes implicated in ADPKD has expanded. Using cohorts of patients who were PKD1 and PKD2 mutation negative, heterozygous missense and nonsense mutations in GANAB, were found to cause ADPKD-like disease with mild kidney involvement but with severe liver disease [5]. In vitro studies indicate that GANAB is required for polycystin-1 (PC1) and polycystin-2 (PC2) maturation and localization at the cilium [5]. Subcellular localization of GANAB and its exact ciliary role need to be confirmed. WES was also performed in two large families with genetically unresolved ADPKD-like disease and identified

heterozygous mutations in DNAJB11. Additional families were solved using targeted sequencing of DNAJB11, with the shared phenotype being non-enlarged cystic kidneys [6].

DNAJB11 is a co-factor of the endoplasmic reticulum chaperone binding immunoglobulin protein (BiP), which regulates the folding and trafficking or degradation of membrane/secretory proteins and was shown to have a fundamental role in the trafficking of PC1 to the cell membrane [6]. Unsolved cohorts of patients with ADPKD-like phenotypes now need to be screened for these novel causes of ADPKD.

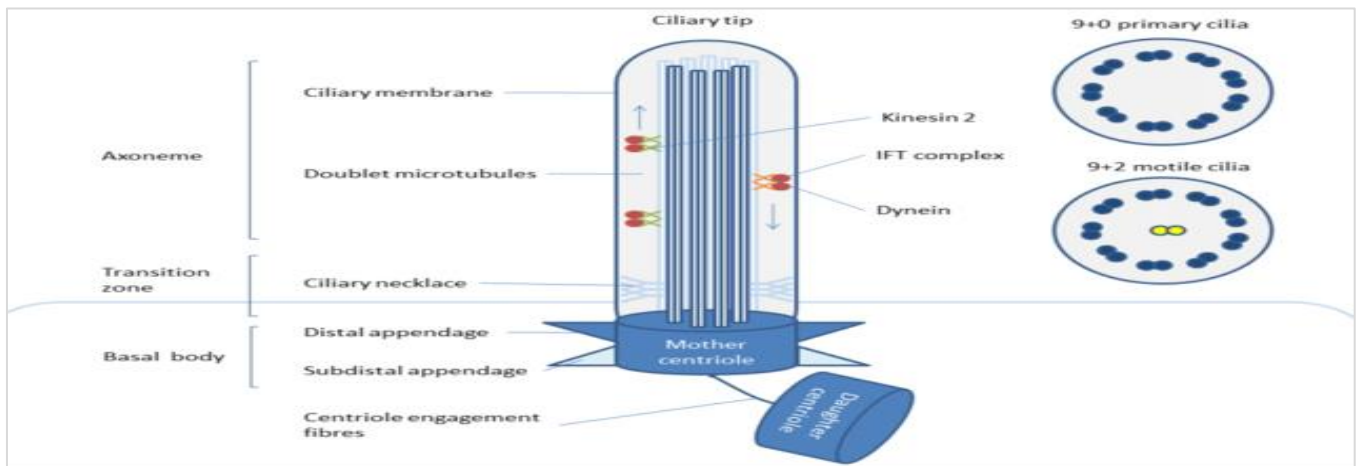


Figure 1: A simplified schematic structure of primary cilia & the cross-sections of cilia axoneme

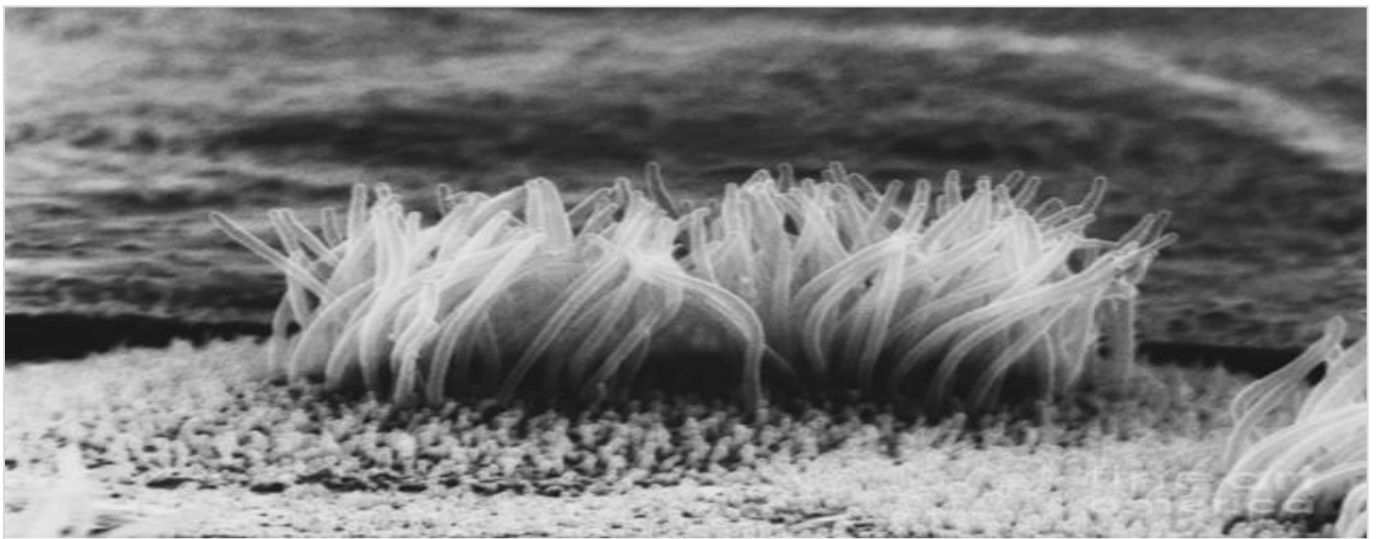
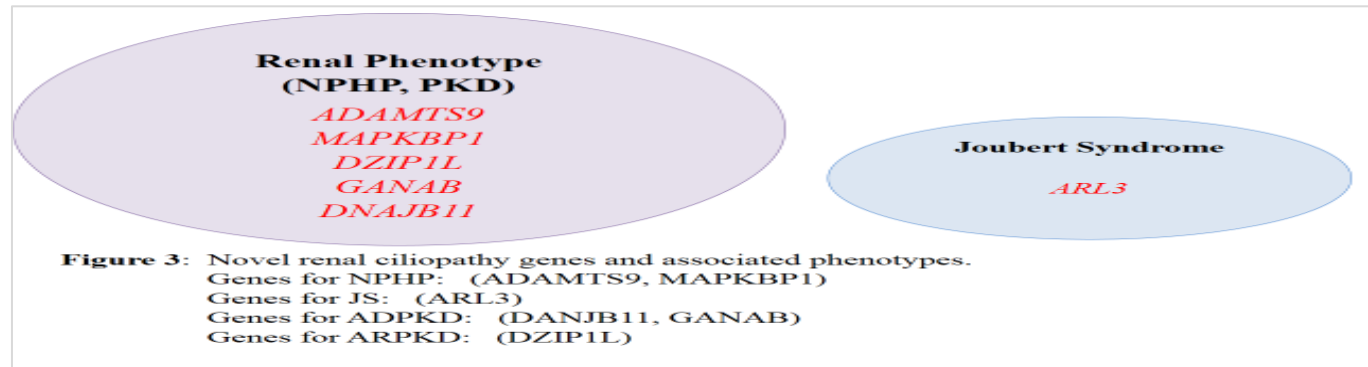


Figure 2: SEM micrograph of motile cilia

Table 1: Summary of novel renal ciliopathy genes

Gene	Encoding protein	Novel patient phenotype
ADAMTS9	A disintegrin and metalloproteinase with thrombospondin type-1 motifs 9	Nephronophthisis-like
MAPKBP1	Mitogen-activated protein kinase binding protein 1	Nephronophthisis-like
ARL3	ADP Ribosylation factor like GTP-ase 3	Joubert Syndrome
DNAJB11	DnaJ heat shock protein family member 11	Autosomal dominant polycystic kidney disease
GANAB	Alpha-Glucosidase II	Autosomal dominant polycystic kidney disease
DZIP1L	DAZ interacting protein 1-like	Autosomal recessive polycystic kidney disease



Novel genes for ADPKD: GANAB and DNAJB11

ADPKD is a well-known and established condition leading to polycystic kidney disease. It accounts for 10% of end-stage renal disease (ESRD). Clinical cohorts have been phenotypically studied in great detail [4]. Over 90% of patients with ADPKD will have mutations in the known genes PKD1 or PKD2, but recently the number of genes implicated in ADPKD has expanded. Using cohorts of patients who were PKD1 and PKD2 mutation negative, heterozygous missense and nonsense mutations in GANAB, were found to cause ADPKD-like disease with mild kidney involvement but with severe liver disease [5]. In vitro studies indicate that GANAB is required for polycystin-1 (PC1) and polycystin-2 (PC2) maturation and localization at the cilium [5]. Subcellular localization of GANAB and its exact ciliary role need to be confirmed. WES was also performed in two large families with genetically unresolved

ADPKD-like disease and identified heterozygous mutations in DNAJB11. Additional families were solved using targeted sequencing of DNAJB11, with the shared phenotype being non-enlarged cystic kidneys [6].

DNAJB11 is a co-factor of the endoplasmic reticulum chaperone binding immunoglobulin protein (BiP), which regulates the folding and trafficking or degradation of membrane/secretory proteins and was shown to have a fundamental role in the trafficking of PC1 to the cell membrane [6]. Unsolved cohorts of patients with ADPKD-like phenotypes now need to be screened for these novel causes of ADPKD.

Novel genes for ARPKD: DZIP1L

ARPKD has been classified as a genetically homogenous disease, caused by recessive mutations in PKHD1 encoding the primary cilia-associated protein fibrocystin. Exact disease mechanisms remain undetermined;

however, fibrocystin is hypothesized to play regulatory roles in primary cilia mechanosensation, calcium signaling, and planar cell polarity through its interaction with PC2. It is also predicted to have multiple ligands, which are vital for cell-matrix and cell-cell interactions [7]. A recent study has indicated that biallelic mutations in DZIP1L is another genetic cause of ARPKD [8]. Lu *et al.* used a combination of genome-wide SNP analysis, WES, and Sanger sequencing in two unrelated consanguineous families, with a total of five affected children, all of them were diagnosed with genetically unsolved ARPKD and identified homozygous missense mutations in DZIP1L. Studies with murine embryonic fibroblasts from *Dzip11^{wpy/wpy}* and human mutant fibroblasts indicated that loss of DZIP1L function contributes to improper distribution of PC1 and PC2 along the ciliary axoneme, with aggregation of both proteins occurring at the tip [8], pointing to an overlap of ADPKD and ARPKD disease mechanism.

Novel genes for Joubert syndrome: ARL3

Joubert syndrome is a multisystem ciliopathy syndrome with AR-inheritance characterized by ataxia (constant finding), hypotonia, global developmental delay, intellectual and physical disability, and oculomotor apraxia. It is diagnosed by the hallmark of molar tooth sign on brain MRI, caused by cerebellar vermis hypoplasia **Figure 4**. It is often associated with retinal degeneration and cystic kidney phenotypes (mainly NPHP) and/or renal cystic dysplasia. Up to date; 34 mutations have

been identified (33 mutations as AR and one mutation as X-linked recessive) [9]. All encoded proteins were related to the structure/function of primary cilium. Recently, ARL3 (ADP-Ribosylation Factor-Like 3) mutations were identified as a novel cause of Joubert Syndrome (JBTS35) [10].

ARL3 encodes a small Ras-related GTP-binding ciliary protein that traffics lipid-modified proteins into the cilium. In a recent study, autozygosity mapping and WES identified two unsolved unrelated consanguineous families as having homozygous missense mutations in ARL3, both resulting in an amino acid substitution at Arg149 [10]. All four affected individuals demonstrated a predominant cerebellar hypoplasia phenotype, as well as retinal abnormalities. One patient presented with left multicystic dysplastic kidney and hydronephrosis. The value of new gene identification is that it allows new disease mechanisms to be explored. Within the primary cilium, ARL3 in its GTP-bound state allows the release of GDI-like Solubilizing factor (GSF) bound ciliary cargo within the cilium. Missense mutations involving the Arg149 residue of ARL3 resulted in the inability of ARL3 to interact with its guanine nucleotide exchange factor ARL13B and therefore led to reduced ciliary cargo release, including NPHP3 [10]. It seems likely that ARL3 mutations may influence downstream ciliary signaling, given the changes seen in ciliary protein composition. A possible therapeutic intervention for ARL3 ciliopathies would include the release of GSF-bound cargo, by small compound, which may compensate for the molecular defect.

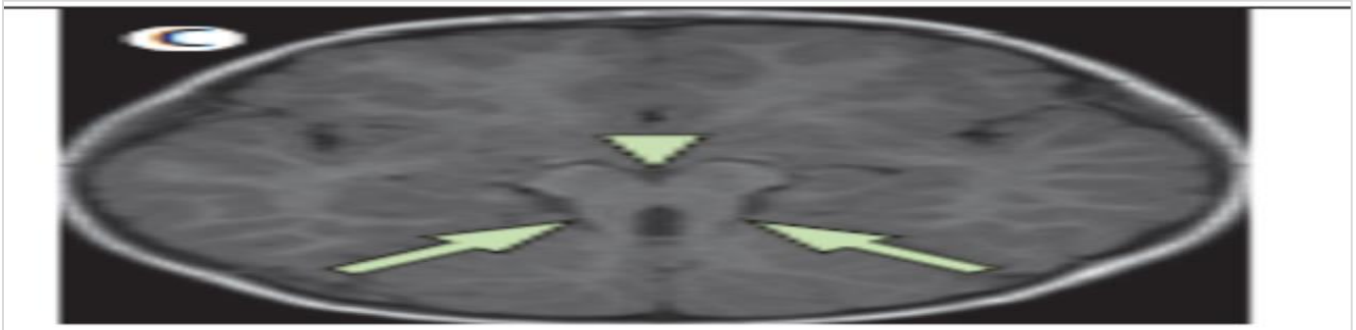


Figure 4: MRI of the brain showing molar tooth sign caused by cerebellar vermis hypoplasia

Novel genes for Nephronophthisis: *ADAMTS9* and *MAPKBP1*

Nephronophthisis is an autosomal recessive cystic kidney disease that progresses to ESRD. It is caused by variants in a large number of genes that encode proteins involved in the function of primary cilia, basal body, and centrosome resulting in renal disease and extrarenal manifestations. UpToDate; more than 20 different genes have been associated with NPHP. The majority of cases show mutations in the NPHP-1 gene [11].

Three clinical variants have been described based on the median age of onset of ESRD:

- Juvenile form: 13 years of age
It is the most common variant and is associated with mutations in all NPHP genes except NPHP-2. The most common affected gene in this type is NPHP-1.
- Infantile form: one year of age
It is associated with mutations in the NPHP-2 gene.
- Adolescent form: 19 years of age
It is associated with mutations in the NPHP-3 gene [11].

Pathology of NPHP

Pathology of NPHP is demonstrated in **Figure 5** shows characteristic corticomedullary cysts in a small scarred kidney. **Figure 6** shows various tubular changes including tubular collapse and thickened tubular basement membrane.

Using a similar methodology in a cohort of patients with NPHP-related ciliopathies, recent studies implicated *ADAMTS9* mutations as a novel renal ciliopathy gene in two individuals with NPHP and early-onset ESRD with various extrarenal manifestations including deafness and short stature [12]. *ADAMTS9* is a metalloproteinase enzyme, vital in the cleavage of ECM proteoglycans and ER-Golgi transportation regulation. Further, in vitro proof of concept studies suggested a role for *ADAMTS9* in ciliogenesis. A zebrafish *adams9* knock down also demonstrated a characteristic ciliopathy phenotype, curved body axis, and hydrocephalus, with renal cysts [12].

Causative compound heterozygous and homozygous mutations in mitogen-activated protein kinase-binding protein 1 (*MAPKBP1*) encoding the non-ciliary scaffold protein have been identified in patients with NPHP. Analysis of patient fibroblasts, with loss of *MAPKBP1*, shows no ciliary phenotype [13].

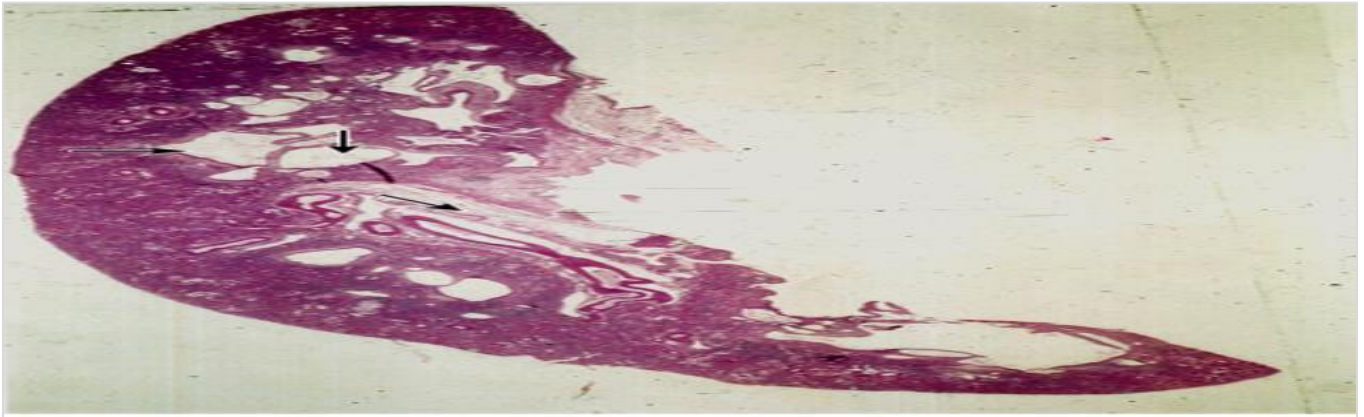


Figure 5: Nephrectomy specimen from a 15-year-old patient with nephronophthisis, demonstrating characteristic corticomedullary cysts (arrows) in a small, scarred kidney.

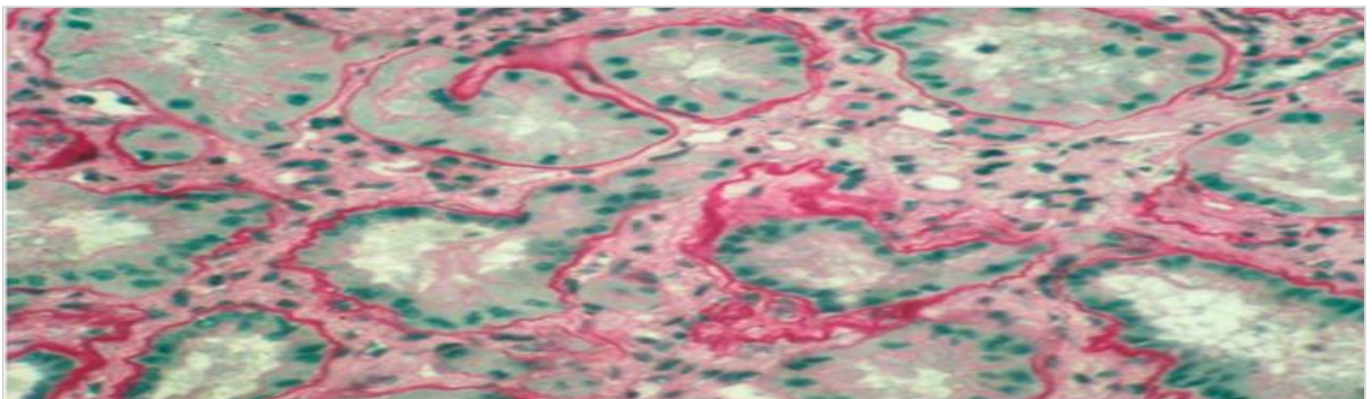


Figure 6: Histological cross section of kidney showing various tubular changes including tubular collapse and thickened tubular basement membranes. Note the laminated and wrinkled appearance of some tubular basement membrane segments as well as the abrupt.

Renal ciliopathy clinical trials

There are currently no curative treatments for renal ciliopathy syndromes, such as NPHP, JS, ARPKD, and ADPKD. There are, however, multiple clinical trials presently ongoing. ADPKD remains an active area of study. Vasopressin receptor inhibitors, such as tolvaptan, continue to be studied to determine their role in slowing cystic kidney growth in ADPKD. Tolvaptan is the first disease-modifying drug to be licensed for the treatment of ADPKD, and practical guidance for its use in clinical practice has recently been published [15]. The use of mTOR inhibitors (sirolimus, everolimus) to retard

cyst growth in polycystic kidney disease has also been reported in clinical trials. The AMPK activator metformin and a multi-tyrosine kinase inhibitor, Bostunib, are part of ongoing studies. Other studies have monitored the effect of reducing protein and sodium intake, alongside water intake, to decrease vasopressin production, as a treatment for ADPKD. A small study demonstrated a significant reduction in serum copeptin from baseline levels in those with a diet and water adjustment compared to controls, indicative of lower vasopressin production, with changes in urinary solutes [16].

CONCLUSIONS

The field of renal ciliopathies is rapidly evolving. With each new gene discovery comes the opportunity to identify novel genetic mechanisms. The use of WES strategies will ensure that genes continue to be identified; however, there is a need to shift towards moving ciliopathy genetics into mainstream medicine so all patients with cystic kidney disease phenotypes can receive, where

possible, a genetic diagnosis. In addition to genetic studies, molecular studies in patient cell lines and model organisms including mice and zebrafish have expanded our insights of the disease mechanisms underlying renal ciliopathies. Current and future challenges involve using these discoveries towards the development of targeted (personalized) therapies to delay and preferably prevent the effects of renal ciliopathy syndrome on the kidneys and other involved organs.

ABBREVIATIONS

ARPKD	Autosomal Recessive Polycystic Kidney Disease
ADPKD	Autosomal Dominant Polycystic Kidney Disease
ADAMTS9	A disintegrin and metalloproteinase with thrombospondin type-1 motifs 9
ARL3	ADP Ribosylation factor like GTP-ase 3
BiP	B immunoglobulin protein
DNAJB11	DnaJ heat shock protein family member 11
DZIP1L	DAZ interacting protein 1-like
ESRD	End Stage Renal Disease
GANAB	Alpha-Glucosidase II
GSF	GDI Like Solubilizing Factor
JS	Joubert Syndrome
NPHP	Nephronophthisis
MAPKBP1	Mitogen Activated Protein Kinase Binding Protein 1
PC1	Polycystin-1
PC2	Polycystin-2
WES	Whole Exome Sequencing

REFERENCES

- Braun DA, Hildebrandt F: Ciliopathies. Cold Spring Harb Perspect Biol 2017 Mar 1; 9(3): a028191. doi: 10.1101/cshperspect.a028191.
- Srivastava S, Molinari E, Raman S, Sayer JA: Many Genes—One Disease? Genetics of Nephronophthisis (NPHP) and NPHP-Associated Disorders. Front. Pediatr. 2017 Sec. Pediatric Nephrology Volume 5, 5:287. <https://doi.org/10.3389/fped.2017.00287>
- Groopman EE, Marasa M, Cameron-Christie S, Petrovski S, Aggarwal VS, Milo-Rasouly H, Li Y, Zhang J, Nestor J, Krithivasan P, et al.: Diagnostic utility of exome sequencing for kidney disease. N Engl J Med 2019, 380:142-151.
- Bae KT, Zhou W, Shen C, Landsittel DP, Wu Z, Tao C, Chapman AB, Torres VE, Yu ASL, Mrug M et al.: Growth pattern of kidney cyst number and volume in autosomal dominant polycystic kidney disease. Clin J Am Soc Nephrol 2019 Jun 7; 14(6):823-833. doi: 10.2215/CJN.10360818.
- 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 Ialpha subunit, cause autosomal-dominant polycystic kidney and liver disease. Am J Hum Genet 2016, 98:1193-1207.
- 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.

7. Israeli S, Amsler K, Zheleznova N, Wilson PD: Abnormalities in focal adhesion complex formation, regulation, and function in human autosomal recessive polycystic kidney disease epithelial cells. *Am J Physiol Cell Physiol* 2010, 298:C831- 46.
8. 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.
9. Agarwal BD, Mohapatra S, Singh S, Guduru V, Nayak S: Neonatal Joubert syndrome with renal involvement and respiratory distress. *Cureus* 2022, 14(5): e24907. doi:10.7759/cureus.24907.
10. Alkanderi S, Molinari E, Shaheen R, Elmaghloob Y, Stephen LA, Sammut V, Ramsbottom SA, Srivastava S, Cairns G, Edwards N et al.: *ARL3* Mutations Cause Joubert Syndrome by Disrupting Ciliary Protein Composition *Am J Hum Genet.* 2018; 103(4):612–620. doi: 10.1016/j.ajhg.2018.08.015.
11. Patrick Niaudet Genetics and pathogenesis of nephronophthisis. *Uptodate* 2023. <https://www.uptodate.com/contents/6136/print>.
12. Choi YJ, Halbritter J, Braun DA, Schueler M, Schapiro D, Rim JH, Nandadasa S, Choi WI, Widmeier E, Shril S et al.: Mutations of *ADAMTS9* cause nephronophthisis-related ciliopathy. *Am J Hum Genet.* 2019 Jan 3; 104(1):45-54. doi: 10.1016/j.ajhg.2018.11.003.
13. Macia MS, Halbritter J, Delous M, Bredrup C, Gutter A, Filhol E, Mellgren AEC, Leh S, Bizet A, Braun DA et al.: Mutations in *MAPKBP1* cause juvenile or late-onset cilia-independent nephronophthisis. *Am J Hum Genet* 2017, 100: 372.
14. Chebib FT, Perrone RD, Chapman AB, Dahl NK, Harris PC, Mrug M, Mustafa RA, Rastogi A, Watnick T, Yu ASL, Torres VE: A practical guide for treatment of rapidly progressive ADPKD with Tolvaptan. *J Am Soc Nephrol* 2018, 29:2458-2470.
15. Amro OW, Paulus JK, Noubary F, Perrone RD: Low-osmolar diet and adjusted water intake for vasopressin reduction in autosomal dominant polycystic kidney disease: a pilot randomized controlled trial. *Am J Kidney Dis* 2016, 68:882-891.

AUTHOR’S CONTRIBUTIONS

The submitted review article is the work of the author.

He has read and approved the manuscript.

Drafting the manuscript by the author.

Revising the manuscript critically for important intellectual content by the author.

Approval of the version of the manuscript to be published by the author.

STATEMENTS

Ethics approval and consent to participate.

This review article was approved and deemed sufficient by the Ethical Committee of Al Azhar University.

Consent for publication

The contents and material of the review article have not been previously reported at any length or being considered for publishing elsewhere.

Availability of data and material

“Not applicable”

Conflict of interest

The author declares no conflict of interest.

Funding

The author declare that this research work did not receive any fund.

Acknowledgements

Author would like to thank all Pediatric nephrology patients and their family members for their valuable contributions to the study.

Submitted: 21/12/2023

Accepted: 27/12/2023

Published online: 31/12/2023