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# Bilateral Perisylvian Polymicrogyria, Intellectual Disability and Nephronophthisis Associated With Compound Heterozygous Pathogenic Variants in the *CEP83* Gene

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**Keywords:** CEP83 | exome sequencing | nephronophthisis | pathogenic variant | polymicrogyria

## ABSTRACT

The centrosomal protein 83 (CEP83) is a centriolar protein involved in primary cilium assembly, an early and critical step in ciliogenesis. Bi-allelic pathogenic variants in the *CEP83* gene have been associated with infantile nephronophthisis and, in a few patients, retinitis pigmentosa. We describe a 5-year-old boy with bilateral perisylvian polymicrogyria, intellectual disability, and nephronophthisis in whom, using exome sequencing, we identified the c.1052T>G p.(Leu351*) stopgain variant inherited from the father and the c.2024T>C p.(Leu675Pro) missense variant inherited from the mother, in a compound heterozygous pattern. Polymicrogyria or, in general, malformations of cortical development had not been previously observed in patients with pathogenic *CEP83* variants. However, defects in CEP83 can affect the formation and function of cilia or centrosomal structures, resulting in a polymicrogyric pattern overlapping with that associated with pathogenic variants affecting other genes coding for centrosomal components. This observation expands the spectrum of phenotypes associated with the *CEP83* gene and adds it to the list of genes associated with bilateral perisylvian polymicrogyria.

## 1 | Introduction

Bi-allelic pathogenic variants in the *CEP83* gene have been associated with nephronophthisis, an autosomal-recessive chronic tubulointerstitial nephritis that generally progresses to end-stage renal disease (ESRD) in childhood (Failler et al. 2014). Infantile nephronophthisis is characterized by hyperechogenic kidney with variable kidney sizes. Some affected individuals also manifest extrarenal symptoms, including intellectual disability, cerebellar ataxia, retinitis pigmentosa, bone abnormalities, or liver fibrosis, defining syndromes referred as nephronophthisis-related ciliopathies (NPHP-RCs) (Failler et al. 2014). In rare patients, pathogenic *CEP83* variants cause retinitis pigmentosa without kidney dysfunction (Veldman et al. 2021).

*CEP83* (NM_016122.3) is located on chromosome 12q22 and encodes the centrosomal protein 83 (CEP83), a 701-residue protein predicted to be mainly composed of coiled-coil domains (Failler et al. 2014) and acts as a component of the distal appendage proteins (DAPs) of the centrioles. DAPs are involved in the anchoring of the mother centriole to the cell membrane, an early and critical step in ciliogenesis (Lo et al. 2019; Joo et al. 2013;

Tanos et al. 2013; Shao et al. 2020; Mansour et al. 2021). CEP83 recruits other DAP components to the ciliary base, and loss of CEP83 disrupts ciliogenesis (Tanos et al. 2013). In radial glial progenitor cells, the major neural progenitor cells that generate neurons and glia in the developing mammalian cerebral cortex, removal of CEP83 disrupts DAP assembly and impairs the anchoring of the centrosome to the apical membrane as well as primary ciliogenesis (Yang et al. 2018; Shao et al. 2020).

Here, we describe a 5-year-old boy harboring compound heterozygous variants in the *CEP83* gene in association with a syndromic phenotype of bilateral perisylvian polymicrogyria, intellectual disability, and nephronophthisis. This finding expands the phenotypic spectrum of *CEP83* gene pathogenic variants to include a malformation of the cerebral cortex.

## 2 | Clinical Report

The proband is a 5-year-old boy who was born at term to non-consanguineous Caucasian parents after a normal pregnancy. At birth, head circumference and length were both within the normal range (33 cm, 10°pc; 47 cm, 20°pc), while weight was below average (2220 g, −2 SD). There were no perinatal complications but hypotonia and difficulty in mouth-feeding were noticed since age 5 months when starting weaning. His motor and speech development were delayed with sitting unsupported at 14 months, walking and babbling at 2 years, first words at 2.5 years (Bayley-III Scale at 2 years 8 months revealed moderate developmental delay with more severely impaired motor skills). Subsequent slow improvements were observed, but gross and fine motor impairment with clumsiness and poor coordination were still present at age 4 years. His expressive language remained poor in spite of regular rehabilitation, including sessions of speech therapy, physiotherapy, and full-time support at school.

Limited dysmorphic features were present, including a slightly elongated face and turricephalic head.

Brain MRI, performed at age 3 years, showed bilateral perisylvian polymicrogyria (Figure 1).

Serial EEGs, recorded since age 3 years showed mild slowing of background activity (6–7 Hz) with no epileptiform discharges.

At age 4 years he was diagnosed with nephronophthisis, causing severe renal failure and requiring peritoneal dialysis while awaiting renal transplantation.

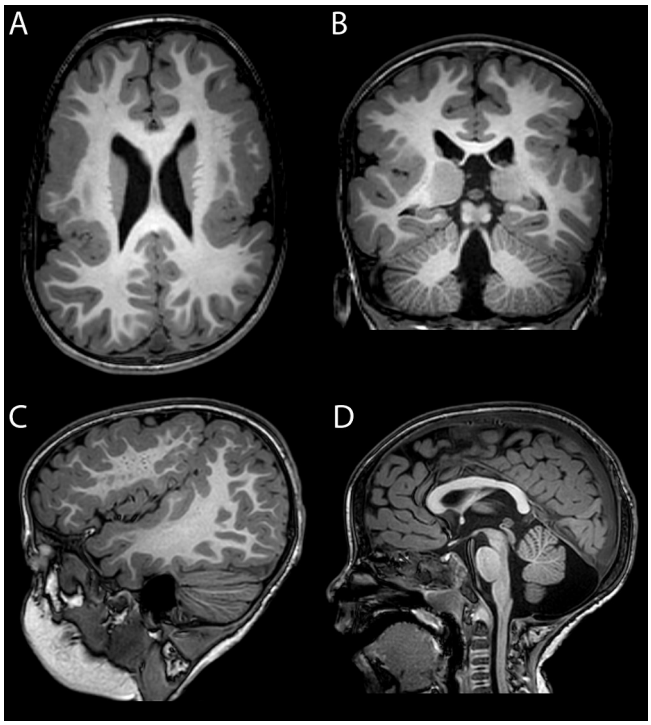


**FIGURE 1** | 3T brain MRI performed when the patient was 3 years old including T1-weighted axial (A), coronal (B), sagittal through the Sylvian fissure (C), and the midline (D) images. The cortical mantle is diffusely abnormal with atypical infolding, packed small thin gyri, and abnormal sulcation. There is more severe involvement of the insulo-sylvian regions, bilaterally, as clearly visible in (A) and (B), but polymicrogyria also extends to the frontal lobes (more on the left hemisphere) and the parieto-temporal regions (more on the right). The lateral ventricles are dysmorphic and mildly asymmetric, with the right slightly larger than the left (A, B). The Sylvian fissure is abnormal with thickened cortex, pronounced infolding and vertically-oriented axis (C). There is dilation of the retrovermian spaces at the level of cisterna magna and some degree of cerebellar vermis atrophy (D).

## 3 | Materials and Methods

Informed consent for exome sequencing (ES) was obtained from the family, and the study was approved by the Pediatric Ethics Committee of the Tuscany Region. Genomic DNA was extracted using a QiaSymphony SP robot (Qiagen, Hilden, Germany), according to the manufacturer's protocol, from peripheral blood leucocytes of the proband and his parents.

We performed ES in the proband—parents trio on exon targets isolated by capture using SureSelect Clinical Research Exome V2 (67.3 Mb) kit (Illumina, San Diego, CA). We sequenced libraries with the Illumina NextSeq 2000 sequencing system with 150 bp paired-end reads (Illumina, San Diego, CA) to an average depth of coverage of at least 70×. For ES data analysis from FASTQ to variant calling, we used the Illumina Dragen Germline analysis platform (Illumina, San Diego, CA). We aligned the reads to human genome build GRCh37/UCSC hg19 and we quality-filtered variants according to GATK's best practices, annotated with VarSeq (Golden Helix, USA), and filtered against public (gnomAD V.2.0) and in-house databases to retain private and rare (MAF <0.1%) variants located in exons with any effect on the coding sequence, and within splice site regions. We filtered variants with depth >10 and genotype quality >20 according to the *de novo* autosomal dominant, X-linked, homozygous recessive, and compound heterozygous models. We evaluated the functional impact of variants through in silico prediction using the dbNSFP database (v3.3a) (Liu et al. 2016). We obtained variants validation and segregation in the family using Sanger sequencing on both strands, with the Big Dye Terminator V3.1 chemistry (Thermo Fisher, MA, USA), on a 3500 DX Genetic Analyzer (Thermo Fisher, MA, USA). We classified variants according to the standard American College of Medical Genetics (ACMG) guidelines

(Richards et al. 2015) and submitted them to the ClinVar database (https://www.ncbi.nlm.nih.gov/clinvar/).

To evaluate the amino acid residue evolutionary conservation, we aligned the CEP83 protein to its orthologues from nine different vertebrates and invertebrate species (Pan troglodytes, Macaca mulatta, Canis lupus familiaris, Bos taurus, Mus musculus, Rattus norvegicus, Gallus gallus, Danio rerio, Xenopus tropicalis) using the Clustal Omega tool (Sievers and Higgins 2018). To evaluate the coiled-coil domains in the CEP83 protein, we analyzed the CEP83 sequence (UniProt Q9Y592) using Waggawagga, a web-based tool for the comparative visualization of coiled-coil predictions and the detection of stable single a-helices domains (Simm, Hatje, and Kollmar 2015).

## 4 | Results

ES revealed a pathogenic variant c.1052T>G p.(Leu351*) and a likely pathogenic variant c.2024T>C p.(Leu675Pro) in the *CEP83* gene (NM_016122.3). The c.1052T>G variant was inherited from the father and the c.2024T>C variant from the mother, configuring a compound heterozygous status. Both variants are novel. The c.2024T>C p.(Leu675Pro) variant is not reported in population databases (Exome Aggregation Consortium, gnomAD database, 1000 Genomes Project and NHLBI Exome Sequencing Project, ESP6500 database), whereas the c.1052T>G p.(Leu351*) variant is reported once in the gnomAD database (1/244,010 alleles; 0 homozygous). According to the American College of Medical Genetics (ACMG) guidelines, the c.1052T>G p.(Leu351*) variant is classified as pathogenic (PVS1, PM2) since it is truncating and predicted to undergo nonsense-mediated RNA decay. The c.2024T>C variant results in the Leu675Pro amino acid substitution of a leucin to proline in the C-terminal coiled-coil domain of the protein that in silico tools (SIFT, Polyphen2, and Mutation Taster) predict to be damaging. The p.Leu675 amino acid residue is highly conserved in the CEP83 human paralogues and in the vertebrates and invertebrates orthologues of the protein (Figure S1); this residue is located in the most carboxy-terminal coiled-coil domain as predicted by three different coiled-coils prediction tools (Marcoils, Ncoils, and Paircoil2) (Figure S2). Despite polymicrogyria was not previously reported in association to *CEP83* defects, we considered the co-occurrence of nephronophthisis and developmental delay in our patient as highly specific for the phenotype associated with bi-allelic *CEP83* variants, thus activating the PP4 ACMG criterion. Therefore, according to ACMG guidelines, we classified the variant as likely pathogenic (PM2, PM3, PP3, and PP4).

## 5 | Discussion

The primary cilium is a membrane-bound structure with a microtubule-based core that originates from the centriole distal end. Ciliogenesis is tightly coupled to the cell cycle and occurs when cells are in G0 or early G1 phase. In the initiating stage, the distal end of the centriole docks to Golgi-derived membrane vesicles. This docking process is mediated by a pinwheel-like structure named the centriole distal appendage (Schmidt et al. 2012; Tanos et al. 2013). CEP83 is a member of the centrosome DAPs involved in distal appendage formation.

Bi-allelic pathogenic *CEP83* variants (OMIM# 615847) have been identified in 11 individuals with early-onset nephronophthisis, an early onset kidney disease that results in end-stage renal disease before the age of 3 years (Failler et al. 2014; Mann et al. 2019; Yue et al. 2020; Stranneheim et al. 2021). In addition to kidney dysfunction, some patients had hydrocephalus, macrocephaly, intellectual disability, strabismus, retinal degeneration, dysmorphisms, cholestasis, and portal fibrosis (Failler et al. 2014; Mann et al. 2019; Stranneheim et al. 2021). Isolated, nonsyndromic retinitis pigmentosa has been reported in four individuals (Haer-Wigman et al. 2017; Veldman et al. 2021; Lynn et al. 2022) (Table S1).

The 5-year-old boy we describe here, carried two compound heterozygous *CEP83* variants (p.Leu351* and p.Leu675Pro) in association with developmental delay, polymicrogyria and nephronophthisis. Brain MRI showed polymicrogyria to be localized in the insular-perisylvian regions bilaterally. Polymicrogyria is a malformation of cortical development characterized by an excessive number of abnormally small gyri that produce an irregular cortical surface with lumpy aspect. The perisylvian cortex is the most frequently affected brain area. Associated clinical manifestations include intellectual disability, impaired oromotor skills and epilepsy (Guerrini and Dobyns 2014). Polymicrogyria can have multiple genetic etiologies or be caused by intrauterine infections, and fetal vascular defects (Parrini et al. 2016). Chromosome abnormalities and pathogenic variants in more than 50 genes coding for tubulin subunits, collagens, centrosome proteins, ion conducting proteins, and components of the PI3K–AKT–mTOR pathway have been causally associated with polymicrogyria (Akula et al. 2023). Genes coding for tubulins and for components of the PI3K–AKT–mTOR pathway have been particularly associated with perisylvian polymicrogyria (Stutterd and Leventer 2014; Mirzaa et al. 2015). Recent evidences indicate that also defects in sodium channels (SCN2A and SCN3A) and sodium potassium pumps (ATP1A2 and ATP1A3) can cause perisylvian polymicrogyria (Smith et al. 2018; Vlachou et al. 2019; Vetro et al. 2021). Hydrocephalus and macrocephaly, observed in one patient each, are the only brain abnormalities reported in association with *CEP83* related nephronophthisis (Failler et al. 2014). However, since only for 2 out of the 15 reported patients harboring bi-allelic variants in this gene brain MRI findings are known, it is possible that polymicrogyria might have been undiagnosed in these patients (Failler et al. 2014; Mann et al. 2019; Yue et al. 2020; Veldman et al. 2021; Stranneheim et al. 2021; Lynn et al. 2022; Haer-Wigman et al. 2017). Although polymicrogyria or, in general, malformations of cortical development, have not been reported in patients with *CEP83* mutations, a link between CEP83 and cortical formation is known. Radial glial progenitors (RGPs) are the major neural progenitors generating neurons and glia in the developing mammalian cerebral cortex. The centrosome in RGPs is anchored to the apical membrane via the DAPs. In mice, removal of Cep83 in RGPs disrupts DAP formation and causes centrosome detachment from the apical membrane, resulting in microtubule disorganization and apical membrane stretching and stiffening. This removal promotes excessive RGPs proliferation and subsequent intermediate progenitor overproduction, generating an enlarged cortex with abnormal folding. Centrosome has a key role in regulating the mechanical features of neural progenitor cells and the size and configuration of the cerebral cortex, making CEP83 an

important player in controlling human brain development and function (Shao et al. 2020). CEP83 is a centriolar protein involved in primary cilium assembly and centrosome formation. Defects in proteins involved in the biogenesis and function of cilia (including DYNC1H1, TMEM161B, and TMEM216), microtubules (including KIF2A, KIF26A, KIF5C, MAST1, TUBA1A, TUBB2B, and TUBG1) and centrosomal structures (including RTTN, ASPM, and WDR62) have all been associated with different types of polymicrogyria (Akula et al. 2023). CEP83 involvement in the same cellular processes as the above polymicrogyria-related proteins, could subtend a common disease mechanism resulting in polymicrogyria.

Eighteen different missense, nonsense, frameshift, and inframe pathogenic variants in the *CEP83* gene have been reported, all transmitted with an autosomal recessive inheritance. Most of these mutations result in amino acid alterations in the coiled-coil domains of the protein (Failler et al. 2014). As for the two variants observed in our patient, p.Leu351* is truncating and predicted to undergo nonsense-mediated RNA decay, whereas Leu675Pro replaces a highly conserved residue in the most carboxy-terminal coiled-coil domain. This domain, encoded by exon 17, plays an important role in CEP83 interaction with its binding partners CEP164 and IFT20 (Failler et al. 2014). Three pathogenic variants (p.Gln692del, p.Glu669Aspfs14, and p.Glu684del), located in proximity of the p.Leu675 residue, have been demonstrated to affect both centrosome localization and interactions with CEP164 and IFT20 (Failler et al. 2014).

It is not possible to establish how the two variants observed in our patient result in the co-occurrence of bilateral perisylvian polymicrogyria with nephronophthisis and intellectual disability. We cannot exclude that the patient's genetic background or a nongenetic cause might have contributed to cortical dysgenesis. A different genetic cause is unlikely, since we analyzed the exome data with an agnostic approach using all the possible inheritance models, without limiting the analysis to the known disease genes. Considering the role of the CEP83 protein in the same cellular processes as other known polymicrogyria-related proteins, it might be possible that the combination of an amorph allele determined by the p.Leu351* stopgain variant with a neomorph allele generated by the p.Leu675Pro missense variant and resulting in a novel or gain-of-function activity not found in the wild-type *CEP83* gene, be responsible of the phenotype observed in our patient. In support of this hypothesis is the observation that none of the previously reported patients with bi-allelic *CEP83* variants harbored a missense substitution in the same coiled-coil domain as in our patient.

Our observation indicates that bi-allelic pathogenic *CEP83* variants cause a syndromic phenotype featuring bilateral perisylvian polymicrogyria, intellectual disability, and nephronophthisis, with the cortical malformation being a main clinical feature. *CEP83* might represent a new candidate gene for polymicrogyria and the spectrum of *CEP83*-related disorders might be wider than previously thought. Additional reports of patients with bi-allelic *CEP83* variants, polymicrogyria and intellectual disability, in addition to nephronophthisis are nevertheless necessary to confirm this association.

### Author Contributions

**Elena Parrini:** data acquisition, conceptualization, writing. **Simona Balestrini:** review and editing. **Domenico Rutigliano:** data acquisition. **Maria Luisa Ricci:** data acquisition. **Davide Mei:** data acquisition, conceptualization, writing. **Renzo Guerrini:** data acquisition, conceptualization, writing, editing, funding acquisition.


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### Ethics Statement

Informed consent for exome sequencing (ES) was obtained from the family and the study was approved by the Pediatric Ethics Committee of the Tuscany Region, Italy in the context of the DESIRE and DECODE-EE projects.

### Conflicts of Interest

The authors declare no conflicts of interest.

### Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

### References


Akula, S. K., A. Y. Chen, J. E. Neil, et al. 2023. "Exome Sequencing and the Identification of New Genes and Shared Mechanisms in Polymicrogyria." *JAMA Neurology* 80: 980–988.

Failler, M., H. Y. Gee, P. Krug, et al. 2014. "Mutations of CEP83 Cause Infantile Nephronophthisis and Intellectual Disability." *American Journal of Human Genetics* 94: 905–914.

Guerrini, R., and W. B. Dobyns. 2014. "Malformations of Cortical Development: Clinical Features and Genetic Causes." *Lancet Neurology* 13: 710–726.

Haer-Wigman, L., W. A. van Zelst-Stams, R. Pfundt, et al. 2017. "Diagnostic Exome Sequencing in 266 Dutch Patients With Visual Impairment." *European Journal of Human Genetics* 25: 591–599.

Joo, K., C. G. Kim, M.-S. Lee, et al. 2013. "CCDC41 is Required for Ciliary Vesicle Docking to the Mother Centriole." *Proceedings of the National Academy of Sciences of the United States of America* 110: 5987–5992.

Liu, X., C. Wu, C. Li, and E. Boerwinkle. 2016. "dbNSFP v3.0: A One-Stop Database of Functional Predictions and Annotations for Human Nonsynonymous and Splice-Site SNVs." *Human Mutation* 37: 235–241.

Lo, C.-H., I.-H. Lin, T. T. Yang, et al. 2019. "Phosphorylation of CEP83 by TTBK2 is Necessary for Cilia Initiation." *Journal of Cell Biology* 218: 3489–3505.

Lynn, J., A. Raney, N. Britton, et al. 2022. "Genetic Diagnosis for 64 Patients With Inherited Retinal Disease." *Genes* 14: 74.

Mann, N., D. A. Braun, K. Amann, et al. 2019. "Whole-Exome Sequencing Enables a Precision Medicine Approach for Kidney Transplant Recipients." *Journal of the American Society of Nephrology* 30: 201–215.

Mansour, F., F. J. Boivin, I. B. Shaheed, M. Schueler, and K. M. Schmidt-Ott. 2021. "The Role of Centrosome Distal Appendage Proteins (DAPs)

in Nephronophthisis and Ciliogenesis." *International Journal of Molecular Sciences* 22: 12253.

Mirzaa, G. M., V. Conti, A. E. Timms, et al. 2015. "Characterisation of Mutations of the Phosphoinositide-3-Kinase Regulatory Subunit, PIK3R2, in Perisylvian Polymicrogyria: A Next-Generation Sequencing Study." *Lancet Neurology* 14: 1182–1195.

Parrini, E., V. Conti, W. B. Dobyns, and R. Guerrini. 2016. "Genetic Basis of Brain Malformations." *Molecular Syndromology* 7: 220–233.

Richards, S., N. Aziz, S. Bale, et al. 2015. "Standards and Guidelines for the Interpretation of Sequence Variants: A Joint Consensus Recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology." *Genetics in Medicine* 17: 405–424.

Schmidt, K. N., S. Kuhns, A. Neuner, B. Hub, H. Zentgraf, and G. Pereira. 2012. "Cep164 Mediates Vesicular Docking to the Mother Centriole During Early Steps of Ciliogenesis." *Journal of Cell Biology* 199: 1083–1101.

Shao, W., J. Yang, M. He, et al. 2020. "Centrosome Anchoring Regulates Progenitor Properties and Cortical Formation." *Nature* 580: 106–112.

Sievers, F., and D. G. Higgins. 2018. "Clustal Omega for Making Accurate Alignments of Many Protein Sequences." *Protein Science* 27: 135–145.

Simm, D., K. Hatje, and M. Kollmar. 2015. "Waggawagga: Comparative Visualization of Coiled-Coil Predictions and Detection of Stable Single α-Helices (SAH Domains)." *Bioinforma* 31: 767–769.

Smith, R. S., C. J. Kenny, V. Ganesh, et al. 2018. "Sodium Channel SCN3A (NaV1.3) Regulation of Human Cerebral Cortical Folding and Oral Motor Development." *Neuron* 99: 905–913.

Stranneheim, H., K. Lagerstedt-Robinson, M. Magnusson, et al. 2021. "Integration of Whole Genome Sequencing Into a Healthcare Setting: High Diagnostic Rates Across Multiple Clinical Entities in 3219 Rare Disease Patients." *Genome Medicine* 13: 40.

Stutterd, C. A., and R. J. Leventer. 2014. "Polymicrogyria: A Common and Heterogeneous Malformation of Cortical Development." *American Journal of Medical Genetics. Part C, Seminars in Medical Genetics* 166C: 227–239.

Tanos, B. E., H.-J. Yang, R. Soni, et al. 2013. "Centriole Distal Appendages Promote Membrane Docking, Leading to Cilia Initiation." *Genes & Development* 27: 163–168.

Veldman, B. C. F., W. F. E. Kuper, M. Lilien, et al. 2021. "Beyond Nephronophthisis: Retinal Dystrophy in the Absence of Kidney Dysfunction in Childhood Expands the Clinical Spectrum of CEP83 Deficiency." *American Journal of Medical Genetics. Part A* 185: 2204–2210.

Vetro, A., H. N. Nielsen, R. Holm, et al. 2021. "ATP1A2- and ATP1A3-Associated Early Profound Epileptic Encephalopathy and Polymicrogyria." *Brain: A Journal of Neurology* 144: 1435–1450.

Vlachou, V., L. Larsen, E. Pavlidou, et al. 2019. "SCN2A Mutation in an Infant With Ohtahara Syndrome and Neuroimaging Findings: Expanding the Phenotype of Neuronal Migration Disorders." *Journal of Genetics* 98: 54.

Yang, T. T., W. M. Chong, W.-J. Wang, et al. 2018. "Super-Resolution Architecture of Mammalian Centriole Distal Appendages Reveals Distinct Blade and Matrix Functional Components." *Nature Communications* 9: 2023.

Yue, Z., H. Lin, M. Li, et al. 2020. "Clinical and Pathological Features and Varied Mutational Spectra of Pathogenic Genes in 55 Chinese Patients With Nephronophthisis." *Clinica Chimica Acta* 506: 136–144.

**Supporting Information**

Additional supporting information can be found online in the Supporting Information section.