An Unusual Presentation of Denys-Drash Syndrome Due to Bigenic Disease

abstract

We report a case of Denys-Drash syndrome (DDS) in a 3-month-old girl presenting with bilateral renal cortical cysts mimicking polycystic kidney disease. Genetic analysis revealed a de novo heterozygous missense mutation c.1186G>A (p.Asp396Asn) in the WT1 gene, confirming the diagnosis of DDS. Because multiple renal cysts have never been reported in DDS, we explored several genes responsible for these renal manifestations, such as HNF-1β, PAX2, PKD1, and PKD2. Remarkably, we identified a heterozygous missense variant c.12439A>G (p.Lys4147Glu) in the PKD1 gene. The same variant was found in the patient’s mother, who had no renal cysts, and in the grandfather, who had several renal cysts. Mutation prediction programs classified the c.12439A>G variant as being “likely pathogenic.” We hypothesize that the severe cystic phenotype in the index patient could be due to the WT1 mutation, enhancing pathogenicity of the “hypomorph” PKD1 allele. A possible role for Wilms tumor suppressor 1 (WT1) in renal cyst development should be considered. From a conceptual point of view, this case shows that an unusual presentation of a known genetic syndrome might point to bigenic inheritance, with unexpected interference of mutated genes causing an uncommon clinical phenotype. Pediatrics 2014;133:e252–e256

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KEY WORDS
Denys-Drash syndrome, WT1, PKD1, diffuse mesangial sclerosis, polycystic kidney disease

ABBREVIATIONS
ADPKD—autosomal dominant polycystic kidney disease
ARPKD—autosomal recessive polycystic kidney disease
DDS—Denys-Drash syndrome
PKD—polycystic kidney disease
WT1—Wilms tumor suppressor 1

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Denys-Drash syndrome (DDS) is characterized by a triad of symptoms: congenital or infantile nephrotic syndrome due to diffuse mesangial sclerosis, male pseudohermaphroditism (present in all 46XY children, but not in 46XX children), and a strong predisposition to develop unilateral or bilateral Wilms tumors and gonadoblastomas.1-3 The syndrome is caused by dominant mutations in the WT1 gene located on chromosome 11p13 and encoding a zinc finger DNA-binding protein, Wilms tumor suppressor 1 (WT1).4 WT1 plays an important role in the development of several organs, including the kidney5 and the urogenital tract.6,7 Until now WT1 mutations have never been associated with renal cysts.

Here we describe a girl with a WT1 mutation, who presented initially with bilateral renal cortical cysts mimicking polycystic kidney disease (PKD), and having a heterozygous missense c.12439A>G variant in the PKD1 gene. A mutation in the PKD1 gene is the most frequent cause of autosomal dominant PKD (ADPKD) in humans and is found in 70% to 74% of patients.8,9 ADPKD is marked by extreme phenotypic variability that is likely to be related to other genetic or allelic factors modulating disease severity.10 Recently, the presence of incompletely penetrant or hypomorphic alleles of the PKD1 gene has been described.11 We suggest that in this case the PKD1 variant is a hypomorphic allele that causes the development of ADPKD in association with the WT1 mutation.

PATIENT PRESENTATION

A female infant presented at 3 months of age because of an incidental finding of proteinuria during investigations for fever of unknown origin. She was the second child of healthy, unrelated parents. The antenatal and neonatal history was unremarkable. On physical examination, discrete edema was found. The patient’s blood pressure was 85/50 mm Hg. Blood tests revealed hypoalbuminemia (23.8 g/L) and normal serum creatinine of 0.37 mg/dL (32.7 μmol/L). Urine protein level was elevated (3.5 g/L). Renal ultrasound revealed enlarged kidneys (right: +2.72 SDs; left: +3.10 SDs) with loss of corticomedullary differentiation and bilateral multiple renal cortical cysts (maximum size: 2.47 cm). Neither family history nor renal ultrasound of both parents (mother: age 37 years; father: age 38 years) revealed the presence of renal disease. The diagnosis of autosomal recessive PKD (ARPKD) was suspected; however, pronounced proteinuria did not fit in the diagnosis. Therefore, kidney and liver biopsies were performed. The liver biopsy showed no abnormalities, excluding ARPKD. Histologic examination of the renal tissue showed diffuse mesangial sclerosis (Fig 1).

At the age of 7 months, an abdominal mass was palpable at the left flank. Ultrasound and abdominal computed tomography revealed a nodular soft tissue mass compatible with Wilms tumor (Fig 2A). The kidneys were still enlarged (right: +3.08 SDs; left: +3.08 SDs), and bilateral cortical cysts were present (Fig 2B). A thorough evaluation, including brain and chest computed tomography, revealed no other abnormalities or metastases. The Wilms tumor was treated by heminephrectomy and chemotherapy (vincristine and actinomycin). On histologic examination, this Wilms tumor was characterized as a stromal type Wilms tumor, stage I. Histologic examination of a cyst (located in the resected specimen but separate from the Wilms tumor) showed 1 layer of flattened epithelial cells, without any evidence of malignancy (Fig 3). After left heminephrectomy, sequential renal ultrasounds still showed multiple cysts that did not undergo remarkable evolution. At 20 months of age, the girl developed acute generalized edema and deterioration of renal function. Treatment with hemodialysis was initiated, and bilateral nephrectomy was performed. Histologic examination of the nephrectomized kidneys revealed diffuse mesangial sclerosis and the presence of multiple cysts. At 3.5 years of age, successful cadaver kidney transplantation was performed.

Genetic testing showed a heterozygous missense mutation c.1186G>A (p. Asp396Asn) in exon 9 of the WT1 gene, previously described in patients with DDS.12 The patient’s karyotype was 46XX, consistent with the normal female phenotype. Neither of the parents were carriers of this mutation. Genetic analyses of PKD2, PAX2, and HNF-1β revealed no abnormalities. Genetic analysis of the open reading frame and splice sites of the PKD1 gene revealed a heterozygous missense variant c.12439A>G (p. Lys4147Glu) in exon 45. The potential pathogenicity of this newly identified missense variant was evaluated by

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**FIGURE 1**
Jones staining of kidney biopsy sample at the age of 5 months, revealing mesangial sclerosis (original magnification ×400).
different mutation prediction software programs (PolyPhen [Available at: http://genetics.bwh.harvard.edu/pph2/], SIFT [Available at: http://sift.jcvi.org], and MutationTaster [Available at: http://www.mutationtaster.org/]) and scored as previously described.13 This variation was classified as being likely pathogenic. The same variant was also found in the 37-year-old mother and in the 65-year-old maternal grandfather. Renal ultrasound did not reveal any renal abnormalities in the mother, but in the maternal grandfather 2 cysts in the left kidney and 5 cysts in the right kidney were found. No abnormalities were found in PKD1 in the patient’s father and in the maternal grandmother (Fig 4).

DISCUSSION

To our knowledge, polycystic kidneys have never been described in patients with DDS. Initially, the working diagnosis in this patient was ADPKD or ARPKD. After kidney biopsy and the development of a Wilms tumor, the diagnosis of DDS became clear: We then suggested that the renal cysts could be part of bilateral cystic Wilms tumors. A cystic Wilms tumor, however, consists of a multilocular cyst with nephroblastoma tissue in the septa and is sharply demarcated from the rest of the kidney parenchyma.14 In our patient, there was no radiologic or histopathological evidence of demarcated lesions, nor was there any histopathological evidence of nephroblastoma tissue in the cyst walls.

Because the cause of renal cyst development remained unclear, we performed genetic analysis of several genes known to cause renal cyst formation. Whereas PKD2, PAX2, and HNF-1β showed no abnormalities, c.12439A>G (p. Lys4147Glu) variant was identified in exon 45 of the PKD1 gene, encoding for polycystin-1.15 This unknown variant is located in the XI transmembrane domain of polycystin-1 next to its cytosolic G-protein binding domain.16 Lysine at position 4147 is a conserved amino acid. To test the potential functional significance of p. Lys4147Glu substitution, we used PolyPhen, SIFT, and MutationTaster prediction software programs. These tests classified this variant as being “likely pathogenic.”

As mentioned above, the same variant was found in the mother who had no renal cysts, and in the maternal grandfather, who had a mild cystic phenotype with 2 cysts in the left kidney and 5 cysts in the right kidney, and normal kidney function.

We hypothesize that c.12439A>G is a hypomorphic allele, associated with mild or no cystic disease on its own but leading to early-onset cystic disease in combination with an additional genetic modifier. Hypomorphic alleles of the Pkd1 gene have initially been described in mice, indicating that lowering Pkd1 gene expression is sufficient to cause cyst formation.17 More recently, incompletely penetrant alleles of PKD1

FIGURE 2
Abdominal computed tomography scan at the age of 7 months. A, Wilms tumor in the lower pole of the left kidney (arrow). B, Bilateral enlarged kidneys with multiple cortical cysts and loss of cortico-medullary differentiation.

FIGURE 3
Hematoxylin-eosin staining of cyst wall at 1 year of age, revealing 1 layer of flattened epithelial cells (arrow marked with *) surrounded by a fibrous border (arrow marked with **) without nephroblastoma tissue (original magnification ×50).
were found in humans with ADPKD and were suggested to be functionally analogous to described murine Pkd1 hypomorphic alleles and to cause the remarkable disease variability within the same family. One incompletely penetrant allele of PKD1 was associated with mild cystic disease, whereas homozygosity led to typical or severe cystic disease, and an incompletely penetrant allele of PKD1 in combination with a pathogenic mutation in PKD1 or PKD2 was associated with early-onset cystic disease.\(^{11}\) Furthermore, the role of HNF-1\(b\) and PKHD1 in modifying disease severity in PKD has been demonstrated.\(^{18}\) However, the combination of an incompletely penetrant allele in the PKD1 gene with a WT1 mutation has not been previously described.

The question remains whether the presence of the WT1 mutation in the index case could enhance the pathogenicity of the c.12439A>G variant. WT1 is a transcription factor initially identified as a Wilms tumor suppressor protein in children with WAGR (Wilms tumor, Aniridia, Genito-urinary anomalies and mental Retardation) syndrome.\(^{19}\) The gene consists of 10 exons; exons 1 to 6 encode a proline-glutamine-rich region that is involved in nuclear localization and transcriptional activation or suppression and exons 7 to 10 encode 4 zinc fingers that are responsible for DNA and RNA binding.\(^{20}\) In the fetal kidney, the WT1 gene is expressed in the metanephric blastema, condensing mesenchyme, renal vesicle, and developing podocytes, whereas in the mature kidney WT1 expression persists only in podocytes.\(^{21}\) By using chromatin from embryonic mouse renal tissue, 1663 genetic targets of WT1 were identified.\(^{22}\) Some of these transcriptional targets can be involved in renal cyst development. PAX2 gene expression is downregulated by WT1, and persistent PAX2 expression has been described in podocytes of patients with DDS.\(^{21}\) Moreover, reduced Ptx2 gene dosage leads to a significant inhibition of renal cyst growth in mice due to increased apoptosis in cystic epithelium.\(^{23}\) Another gene that might be interesting in this regard is CTGF, encoding connective tissue growth factor, which is also suppressed by WT1.\(^{12}\) Connective tissue growth factor has been shown to be upregulated in renal cystic epithelia in a Pkd1 mouse model.\(^{24}\) Nab2, which is also a WT1 target gene, can be another possible link between WT1 mutation and renal cyst development. Nab2, encoding nerve growth factor-induced protein A-binding protein-2, is upregulated in vitro by curcumin\(^{25}\); curcumin inhibits renal cyst formation and enlargement in vitro in Madin-Darby canine kidney cells exposed to foskolin\(^{26}\) and in vivo in a PKD-deleted mouse model.\(^{27}\)

Although we provide no mechanistic proof, our case is the first indication that WT1 could be involved in renal cyst formation. Thus, the role of WT1 in renal cystogenesis should be further studied.

From a conceptual point of view, this case shows that an unusual presentation of a known genetic syndrome can point to bigenic inheritance modulating expressivity of mutated genes causing an uncommon clinical phenotype. This phenomenon has recently been described in other genetic disorders such as Dent disease caused by coinheritance of mutations in CLCN5 and OCRL genes.\(^{28}\) The wide use of genome-broad genetic testing is likely to reveal more cases of bigenic disease underlying complex clinical phenotypes in the future.

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