Spectrum of *HNF1B* Mutations in a Large Cohort of Patients Who Harbor Renal Diseases

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Background and objectives: Hepatocyte nuclear factor 1β (HNF1β) is a transcription factor that is critical for the development of kidney and pancreas. In humans, mutations in *HNF1B* lead to congenital anomalies of the kidney and urinary tract, pancreas atrophy, and maturity-onset diabetes of the young type 5 and genital malformations.

Design, setting, participants, & measurements: We report *HNF1B* screening in a cohort of 377 unrelated cases with various kidney phenotypes (hyperechogenic kidneys with size not more than +3 SD, multicystic kidney disease, renal agenesis, renal hypoplasia, cystic dysplasia, or hyperuricemic tubulointerstitial nephropathy not associated with *UMOD* mutation).

Results: We found a heterozygous mutation in 75 (19.9%) index cases, consisting of a deletion of the whole gene in 42, deletion of one exon in one, and small mutations in 32. Eighteen mutations were novel. *De novo* mutations accounted for 66% of deletions and 40% of small mutations. In patients who carried *HNF1B* mutation and for whom we were able to study prenatal ultrasonography (56 probands), isolated hyperechogenic kidneys with normal or slightly enhanced size were the more frequent (34 of 56) phenotype before birth. Various other prenatal renal phenotypes were associated with *HNF1B* mutations, at a lesser frequency. Diabetes developed in four probands. Hyperuricemia and hypomagnesemia, although not systematically investigated, were frequently associated.

Conclusions: This large series showed that the severity of the renal disease associated with *HNF1B* mutations was extremely variable (from prenatal renal failure to normal renal function in adulthood) and was not correlated with the genotype.

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Hepatocyte nuclear factor 1β gene (*HNF1B*) encodes a transcription factor that binds DNA as homodimer or as heterodimer with the related factor HNF1α. Heterozygous mutations of *HNF1B* were first described in maturity-onset diabetes of the young type 5 (1). Renal manifestations are frequently observed in patients with maturity-onset diabetes of the young type 5 and include a wide spectrum of phenotypes (2). More recently, *HNF1B* mutations were found to be associated with a subset of fetal bilateral hyperechogenic kidneys (3) and other kidney diseases diagnosed before birth (4). Besides diabetes, nonrenal anomalies involving Mullerian and Wolffian derivatives, liver and pancreas abnormalities, hyperuricemia with or without gout (5), and hypomagnesemia (6) have been reported.

HNF1B plays a crucial role in early development (7) and thereafter is involved in the organogenesis of several tissues, such as gut, pancreas, liver, lung, and kidney. The gene is also transiently expressed in the neural tube and in the epididymis, vas deferens, seminal vesicle, prostate, uterus, and oviduct (7,8). During kidney development, the gene is expressed in the ureteric bud, in the comma- and S-shaped bodies, and then in the proximal and distal tubules but not in the glomerulus (9). Kidney-specific inactivation of *Hnf1B* in the mouse leads to cystic disease, and HNF1β was shown to bind directly DNA elements that regulate the expression of genes whose mutations are responsible for cystic kidney diseases (*Nphp1*, *polaris*, *Umod*, *Pkhd1*, and *Pkd2*) (10) or of a gene identified as a candidate modifier in a mouse model of cystic kidney disease (*Kif12*) (11). Here we report on *HNF1B* mutation screening in a series of 377 unrelated patients who presented with various kidney phenotypes, giving special attention to the prenatal renal phenotypes.

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Materials and Methods

Patients

This is a retrospective study in which we included all cases that were not previously reported and were tested for *HNF1B* mutations in two reference centers for rare kidney diseases in France. Criteria for inclusion were hyperechogenic kidneys (but with size not more than +3 SD), uni- or bilateral multicystic kidney disease (MCD), renal agenesis, renal hypoplasia, cystic dysplasia, or hyperuricemic tubulointerstitial nephropathy not associated with *UMOD* mutation. Patients' samples, medical records, genealogy, and written informed consent from patient and/or parents were sent from Pediatric, Pediatric Nephrology, Nephrology, or Obstetric Departments. Genomic DNA was extracted from venous blood or tissues collected from 377 unrelated cases (271 children, 57 adults, and 49 fetuses), 221 male and 156 female.

Prenatal ultrasonographs were available for 245 probands (usually performed at 12, 22, and 32 weeks of amenorrhea) and had been considered as normal in only 11 cases. Renal phenotypes before birth were isolated hyperechogenic kidneys (not larger than +3 SD in size) in 55 cases, bilateral MCD (13 cases), unilateral MCD (74 cases), unilateral agenesis (34 cases), bilateral agenesis (13 cases), renal hypoplasia (25 cases), urinary tract dilation (11 cases), and cystic disease (nine cases). In 132 patients, either the result of the prenatal ultrasound was not known or ultrasound was not performed (patients born before 1980). Renal phenotypes after birth were hyperechogenic kidneys (23 cases), unilateral MCD (12 cases), unilateral agenesis (8 cases), renal hypoplasia (33 cases), urinary tract dilation (2 cases), hyperuricemic tubulointerstitial nephritis (18 cases), unclassified cystic disease (35 cases), and only extrarenal symptoms (diabetes and uterine abnormalities; one case).

Patients with renal cavity dilation and/or recurrent acute pyelonephritis had voiding cystourethrogram. GFR was estimated by the Modification of Diet in Renal Disease (MDRD) formula for adults and by the Schwartz formula for children who were younger than 16.

Molecular Analysis

Quantitative multiplex PCR amplification of short fluorescence fragments (12) was performed as described previously (13) for the search of deletion. When deletion was not found, the nine exons and the exon–intron boundaries of the gene were screened for mutations by direct sequencing as described previously (1).

Statistical Analysis

Testing for difference in proportions was performed using the χ^2. All tests were two sided. $P < 0.05$ was considered significant.

Results

Mutations

Heterozygous *HNF1B* alterations, which are thought to be pathogenic, were found in 75 probands (41 male and 34 female), leading to a mutation detection rate of 19.9% of tested index cases. They consisted of a heterozygous deletion of the entire gene in 42 cases (Table 1). Parent status was studied for 21 probands: deletions were *de novo* in 14 of 21 cases and inherited in seven of 21. Mutations that were not deletions of the entire gene are shown in Table 2. One patient was carrying a *de novo* heterozygous deletion of exon 4, which was previously reported (3,13). Twenty-four different heterozygous small mutations (11 missense, five nonsense, five frameshift, and three splice site mutations) were found in 32 probands. Parent status was studied for 20 of them. Mutation were shown to be *de novo* in eight of 20 cases and to be inherited in 12 of 20 cases; 18 were novel. Except for the mutation affecting the initiator codon, all missense mutations were localized in the DNA binding domain (Figure 1), were modifying a conserved amino acid, and were predicted to be probably damaging by the Polyphen program (14). In some families, there was a father-to-son transmission, in agreement with an autosomal dominant mode of inheritance (see proband 64 as an example).

Renal and Extrarenal Phenotype

Patients for Whom Prenatal Ultrasound Was Available

In 245 cases tested for *HNF1B* mutation, we were able to go back to the prenatal ultrasound. Mutations were identified in 56 of them.

Prenatal phenotype in patients with *HNF1B* mutation was isolated bilateral hyperechogenic kidneys with normal or moderately enlarged size in 34 cases, including one termination of pregnancy (TOP) because of an associated oligo-anamnios. Evaluation of these patients at last follow-up showed renal failure with GFR <80 ml/min per 1.73 m^2 (range 32 to 61 ml/min per 1.73 m^2) in eight patients (1 months to 14 years old), GFR >80 ml/min per 1.73 m^2 in 20 patients (1 to 17 years old), and unknown in five patients. Five patients experienced transitory renal failure at birth, and one developed diabetes at the age of 17.

Other prenatal phenotypes in patients with *HNF1B* mutation were bilateral MCD (leading to TOP) in two patients, unilateral MCD in eight patients, unilateral renal agenesis (with hypoplasia and/or cysts on the single kidney) in four patients, unilateral renal hypoplasia in one patient, renal macrocysts in three patients (with urinary tract dilation, pancreas hypoplasia, and TOP in one patient), and isolated upper urinary tract dilation in one patient (who developed small cortical cysts after birth). In three patients who presented with severe cystic dysplasia on early ultrasound, the prenatal ultrasounds were considered as normal. In all cases with unilateral MCD, patients developed postnatal anomalies on the contralateral kidney. In the case with unilateral hypoplasia, cysts developed on the hypoplastic kidney after birth

Patients for Whom Prenatal Ultrasound Was not Available

In 132 patients who were tested for *HNF1B* mutation, we were not able to go back to prenatal ultrasound (either the result of it was not known, or ultrasound was not performed). We found an *HNF1B* mutation in 19 of them, including 10 who were tested during adulthood, six of whom had a family history of renal diseases. Four adult probands had cystic renal hypoplasia (associated with hypomagnesemia, gout, and a diabetes that occurred at 42 years in one and with gestational diabetes in another). Two had hyperechogenic kidneys with microcysts. One had solitary kidney and early gout, and another one had hyperuricemic interstitial nephropathy. One female born from consanguineous parents developed unclassified renal cystic dysplasia with uterus agenesis, imperforated vagina, cleft palate, and mental retardation. One presented with diabetes at the age of 31 and bicornuate uterus. Four adults (aged 29 to 35 years) had normal renal function and five (aged 28 to 33 years) had reduced GFR (65 ml/min per 1.73 m^2

Table 1. Phenotypes in probands with complete *HNF1B* deletions

Probands	Prenatal Renal Phenotype	Postnatal Renal Phenotype	Deletion Inheritance
1	Normal ultrasound	Large kidneys with numerous bilateral cysts, preterminal renal failure at 8 months; father with renal hypoplasia, GFR unknown	ND (parents not tested)
2	Normal ultrasound	Hyperechogenic and cystic kidneys, normal GFR at 6 years	*De novo*
3	Bilateral hyperechogenic kidneys	Hyperechogenic, normal-sized kidneys, normal GFR at 10 months	ND (parents not tested)
4	Bilateral hyperechogenic kidneys	Hyperechogenic kidneys, multiple microcysts, CRF (GFR 29 at 6 years); mother with renal cysts.	Deletion in the mother
5	Bilateral hyperechogenic kidneys	Hyperechogenic, normal-sized kidneys, microcysts, normal GFR at 5 years	ND (parents not tested)
6	Unilateral hypoplasia	Unilateral hypoplasia with cysts, normal GFR at 2 years Father and paternal grandmother with renal cysts	*De novo*
7	Bilateral hyperechogenic kidneys	Small cystic kidneys, normal GFR at 3 years	ND (parents not tested)
8	Bilateral hyperechogenic kidneys	Normal-sized kidney with cortical cysts + left PUJO hyperuricemia, normal GFR at 5 years	ND (parents not tested)
9	Unilateral MCD, contralateral cysts	Unilateral MCD, cortical cysts on contralateral kidney, hyperuricemia, elevated liver enzymes, normal GFR at 10 years	ND (parents not tested)
10	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, normal-sized kidney, normal GFR at 17 months; father with renal hypodysplasia, GFR unknown; paternal grandfather has CRF; previous TOP for MCD and anamnios in the mother	ND (parents not tested)
11	ND	Diabetes, bicornuate uterus TOP in the past because of anamnios, normal GFR at adult age; diabetes in sisters and father	Deletion in the father
12	Bilateral hyperechogenic kidneys	Few cysts, unknown GFR	*De novo*
13	Unilateral MCD, contralateral hyperechogenic kidney	Cysts in the single kidney, normal GFR at 9 years	ND (parents not tested)
14	Bilateral hyperechogenic kidneys, one cortical cyst	Hyperechogenic large (+2 SD) kidneys, CRF (unknown GFR) at 1 month	*De novo*
15	ND	Cystic kidney disease, uterine agenesis, imperforated vagina, mental retardation, normal GFR at 29 years	ND (parents not tested)
16	Bilateral pelvic dilation	Bilateral PUJO, unilateral small cortical cysts, normal GFR at 14 months	*De novo*
17	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, normal GFR at 3 years; brother with pelvic kidney and PUJO; mother with normal kidneys and normal GFR, left hepatic agenesis, pancreas head hypoplasia, bicornuate uterus	Deletion in the mother

Table 1. continued

Probands	Prenatal Renal Phenotype	Postnatal Renal Phenotype	Deletion Inheritance
18	Bilateral hyperechogenic kidneys	Few cysts, normal-sized hyperechogenic kidneys, neonatal renal failure, normal GFR at 20 months; mother with cysts and gestational diabetes	Deletion in the mother
19	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, normal GFR at 5 years; mother with renal cysts and severe cholestasis	Deletion in the mother
20	ND	Bilateral cysts, normal GFR at 11 years	ND (parents not tested)
21	Bilateral hyperechogenic kidneys, cortical cysts (MRI), diaphragmatic hernia	Dedifferentiated kidneys (54 and 58 mm) with cysts, acute renal failure at birth, GFR 40 ml/min per 1.73 m^2 at 2 months	*De novo*
22	Bilateral hyperechogenic kidneys, cortical cysts, oligoamnios	TOP; renal histology showed cystic dilation of nearly all glomeruli with collapsed floculus, glomerular cysts were lined by fibrosis, interstitial fibrosis with rarefied tubules	*De novo*
23	ND	Bilateral hyperechogenic kidneys, cortical microcysts, normal GFR at 17 years	*De novo*
24	ND	Bilateral cortical microcysts, bicornuate uterus, diabetes, normal GFR at 20 years	ND (parents not tested)
25	ND	Bilateral hyperechogenic kidneys, cortical microcysts, normal GFR at 3 years	ND (parents not tested)
26	Bilateral hyperechogenic kidneys, pelvic dilation	Bilateral hyperechogenic hypoplastic kidneys, unknown GFR, microcysts in mother	Deletion in the mother
27	ND	Bilateral hyperechogenic kidneys, cortical microcysts, CRF (GFR 65 at 30 years)	ND (parents not tested)
28	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic kidneys CRF (GFR 40 at 3 years)	ND (parents not tested)
29	ND	Bilateral hyperechogenic kidneys cortical microcysts, normal GFR at 35 years; mother with type 2 diabetes	ND (parents not tested)
30	ND	Bilateral hyperechogenic kidneys, cortical microcysts, normal GFR at 6 years; microcystic sole kidney in mother	ND (parents not tested)
31	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic kidneys, CRF (GFR 35 at 1 year)	ND (parents not tested)
32	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic kidneys, diabetes at 17 years, normal GFR at 20 years	ND (parents not tested)
33	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic kidneys, normal GFR at 1 year	*De novo*
34	Unilateral MCD, other kidney hyperechogenic	Unilateral MCD, other kidney hyperechogenic with pelvic dilation, normal GFR at 3 years	*De novo*
35	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic kidneys, normal GFR at 15 years	*De novo*

Table 1. continued

Probands	Prenatal Renal Phenotype	Postnatal Renal Phenotype	Deletion Inheritance
36	Bilateral hyperechogenic kidneys + unilateral macrocysts	Bilateral hyperechogenic kidneys + unilateral macrocysts, CRF (GFR 55 at 3 years)	*De novo*
37	Unilateral MCD, other kidney hyperechogenic	Unilateral MCD, hyperechogenic kidney, normal GFR at 6 years	*De novo*
38	Bilateral hyperechogenic kidneys, cortical microcysts	Bilateral hyperechogenic kidneys, cortical microcysts, normal GFR at 6 years	*De novo*
39	Unilateral agenesis	Single hyperechogenic kidney, cortical microcysts, normal GFR at 10 years	ND (parents not tested)
40	Unilateral agenesis, hyperechogenic kidney with microcysts	Single hyperechogenic kidney, microcysts, CRF (GFR 23 at 1 year); single kidney with cysts in the mother (GFR 75 at 30 years)	Deletion in the mother
41	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic kidneys, unilateral VUR, unknown GFR	ND (parents not tested)
42	ND	Bilateral hyperechogenic kidneys, cortical microcysts, normal GFR at 3 years	ND (parents not tested)

CRF, chronic renal failure; MRI, magnetic resonance imaging; ND, not done; PUJO, pelvi-ureteric junction obstruction; VUR, vesicoureteral reflux.

to end-stage renal failure), and renal function was unknown for one.

We found an *HNF1B* mutation in nine patients who were tested during childhood, two of whom had a family history of renal disease. Eight probands had hyperechogenic kidney and cysts and one hypoplastic kidney and/or uterus anomalies (n = 2) and/or pancreatic hypoplasia (n = 1). One developed diabetes at the age of 20 years. Renal function was normal in six patients (aged 3 to 20 years) and altered three times (aged 4 to 15 years).

Genotype–Phenotype Correlation

The severity of the renal disease that is associated with *HNF1B* mutation was extremely variable (from prenatal severe renal failure to normal renal function in adulthood). The type of mutation (deletion of the whole gene; missense mutation; or truncating mutation because of nonsense, frameshift, or splice mutation) was analyzed according to the renal phenotype for the 75 probands who carried an *HNF1B* mutation, as well as for other affected family members when their kidney phenotype was known (Figure 2). The percentage of each type of mutation was not statistically different when the group of patients who had prenatal hyperechogenic kidneys was compared with a group that included all other patients. We also looked for a relation between the type of mutation and the severity of the disease in terms of renal failure, independent of the type of renal disease. The patients with severe and early renal failure (six patients with TOP for oligohydramnios and six patients with terminal or preterminal renal failure that occurred before the age of 4 years) were associated either with deletions (seven patients), truncating mutation (three patients), or missense mutations (two patients), a figure that is not different from the proportion of each type of mutation in all patients. Figure 3 shows the number of patients with and without renal failure for each type of mutation. The proportion of patients with renal failure at last follow-up was significantly (P = 0.012) higher in patients who carried a truncating mutation than in patients who carried an *HNF1B* deletion; however, for unknown reasons, patients with truncating mutation were older than patients with gene deletion at last follow-up. This age difference may account, at least in part, for the different severity of the renal failure.

Discussion

To our knowledge, we report here the largest series of phenotypic and genetic analysis of patients who harbor renal diseases that are associated with *HNF1B* mutations. We screened 377 unrelated patients and identified an *HNF1B* mutation or deletion in 75 unrelated cases: 10 adults and 65 children or fetuses. This rate of mutation (19.9%) is not significantly different from that (23%) recently reported in a smaller cohort of children with renal malformation (6). Going back to the prenatal ultrasound when available, we report the renal phenotypes before birth in patients with *HNF1B* mutation and analyzed the evolution of their renal function.

We had information regarding prenatal ultrasound for 245 patients, and this study confirms our previous finding that

Table 2. Mutations and phenotypes in patients with *HNF1B* mutations that are not complete deletions

Probands	Nucleotide Change	Protein Change	Exon (Intron)	Reference	Prenatal Renal Phenotype	Postnatal Renal Phenotype	Mutation Inheritance
43	c.3G→A	p.Met1Ile	1	This study	Bilateral cortical cysts	Bilateral cortical microcysts, normal GFR at 7 years; father with renal cysts	Mutation in the father
44	c.3G→A	p.Met1Ile			Bilateral cortical cysts	Bilateral cortical microcysts, normal GFR at 7 years; father with diabetes and renal cysts	Mutation in the father
45	c.211 delAAGGGCC	p.Lys71fs	1	This study	ND	Hypodysplastic kidneys with microcysts, GFR 45 at 28 years; father with hyperuricemic nephropathy (GFR unknown)	Mutation in the father
46	c.232G→T	p.Glu78X	1	This study	Normal ultrasound	Bilateral cortical cysts, ESRF at 3 months	*De novo*
47	c.232G→T	p.Glu78X	1		MCD ×2	TOP; septated uterus	ND
48	c.322 delG	p.Ala108fs	1	This study	ND	Hyperuricemic nephropathy, ESRF at 33 years; father with hyperuricemic nephropathy (with kidney graft) and diabetes	Mutation in the father
49	IVS1 345–1G→A		(1)	This study	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, neonatal renal failure, normal GFR at 8 years	ND
50	IVS1 345–1G→A		(1)		Unilateral agenesis + hyperechogenic kidney	Single hyperechogenic kidney + CRF (GFR 25 at 17 years)	ND (parents not tested)
51	c.452C→G	p.Ser151Cys	2	This study	Unilateral MCD	Unilateral MCD and cortical cysts on the other kidney, normal GFR at 6 years	ND
52	c.476C→T	p.Pro159Leu	2	This study	Bilateral hyperechogenic kidneys	Isolated hyperechogenic kidneys, ultrasound normalized (size and echogenicity) at 10 months, normal GFR at 10 months	ND

Table 2. continued

Probands	Nucleotide Change	Protein Change	Exon (Intron)	Reference	Prenatal Renal Phenotype	Postnatal Renal Phenotype	Mutation Inheritance
53	c.494 G→A	p.Arg165His	2	(20)	ND	Small hyperechogenic kidneys, CRF (GFR 16 at 4 years); father with renal failure and diabetes	ND
54	c.494G→C	p.Arg165Pro	2	This study	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic hypoplastic kidneys CRF (GFR 32 at 10 years) + pancreatic hypoplasia	*De novo*
55	c.513G→A	p.Trp171X	2	This study	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, normal GFR at 11 months	ND
56	IVS2 544 + 3delAAGT		(2)	(6)	ND	Renal cysts and VUR, normal GFR at 6 years; mother with unilateral cysts and gestational diabetes, normal GFR at 30 years	Mutation in the mother
57	IVS2 544 + 3delAAGT		(2)		ND	Single kidney, gout, CRF (GFR 25 at 65 years); mother and maternal cousin with renal failure; daughter with single kidney	ND
58	IVS2 544 + 3delAAGT		(2)		ND	Cysts, CRF (GFR 60 at 33 years), diabetes, hyperuricemia, elevated liver enzymes, hypomagnesaemia; father with ESRF	ND
59	c.544C→T	p.Gln182X	2	(20)	ND	Hyperechogenic kidneys, CRF (but unknown GFR); diabetes in the mother.	Mutation in the mother
60	c.544C→T	p.Gln182X	2		Unilateral MCD, other kidney with cysts	Unilateral MCD, other kidney with cortical cysts, normal GFR at 3 months	ND
61	c.544C→T	p.Gln182X	2		Unilateral agenesis	Single hypoplastic hyperechogenic kidney, CRF (GFR 55 at 7 years)	ND

Table 2. continued

Probands	Nucleotide Change	Protein Change	Exon (Intron)	Reference	Prenatal Renal Phenotype	Postnatal Renal Phenotype	Mutation Inheritance
62	c.758A→C	p.Gln253Pro	3	(3)	Bilateral MCD	TOP cysts in mother (GFR 62 at 25 years) and grandmother	Mutation in the mother
63	c.717delG	p.Ser242fs	3	This study	Bilateral hyperechogenic kidneys	Hyperechogenic kidneys, cortical microcysts, normal GFR at 1.5 years; father with renal cysts and diabetes	Mutation in the father
64	c.766C→T	p.Pro256Ser	3	This study	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, normal-sized kidneys, normal GFR at 8 years; phenotype in the father unknown	Mutation in the father
65	IVS3 809 + 1G→A		(3)	This study	Bilateral hyperechogenic kidneys, bilateral cysts	Bilateral cortical cysts, unilateral UPJ, neonatal renal failure, normal GFR at 3 years; family history of diabetes; phenotype in the father unknown	Mutation in the father
66	c.840delC	p.Pro280fs	4	This study	Enlarged kidneys, large cysts, pyelic dilation, duplicity, pancreas hypoplasia	TOP	ND
67	c.854G→A	p.Gly285Asp		(17)	Bilateral hyperechogenic kidneys	Bilateral hyperechogenic kidneys + unilateral cortical microcysts, normal GFR at 3 years; mother with renal cysts, GFR 55 at 35 years	Mutation in the mother
68	c.883C→T	p.Arg295Cys	4	(17)	ND	Small and cystic kidneys (unknown GFR); mother with renal cysts and CRF (precise GFR unknown)	Mutation in the mother
69	c.883C→T	p.Arg295Cys	4		Bilateral hyperechogenic kidneys, bilateral cortical cysts	Cortical cysts, hyperuricemia, neonatal renal failure, normal GFR at 8 years	*De novo*

Table 2. continued

Probands	Nucleotide Change	Protein Change	Exon (Intron)	Reference	Prenatal Renal Phenotype	Postnatal Renal Phenotype	Mutation Inheritance
70	c.895T→G	pTrp299Gly	4	This study	Bilateral hyperechogenic kidneys, bilateral cortical microcysts	Bilateral hyperechogenic kidneys, cortical microcysts CRF (GFR 51 at 3 years)	ND
71	c.766C→T	p.Asn302Lys	4	This study	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, CRF (GFR 60 at 7 years)	ND
72	Exon 4 deletion c.810_1045 del236	p.Arg270fs	4	(3,13)	Unilateral MCD	Absence of hypertrophy of the contralateral kidney, VUR, CRF (GFR 65 at 4 years)	*De novo*
73	c.1136C→A	p.Ser379X	5	This study	ND	Hyperechogenic kidneys, cortical microcysts, CRF (GFR 61 at 15 years), didelphic uterus + pancreatic hypoplasia	*De novo*
74	c.1360C→T	p.Gln454X	7	This study	Unilateral MCD, other kidney hyperechogenic	Unilateral MCD, other kidney hyperechogenic with cortical microcysts, normal GFR at 2 years	*De novo*
75	c.delAG1363–1364	p.Ser455fs	7	This study	Bilateral hyperechogenic kidneys	Bilateral cortical cysts, left hypoplastic kidney CRF (GFR 80 at 14 years)	ND

CRF, chronic renal failure; ESRF, end-stage renal failure; UPJ, ureteropelvic junction.

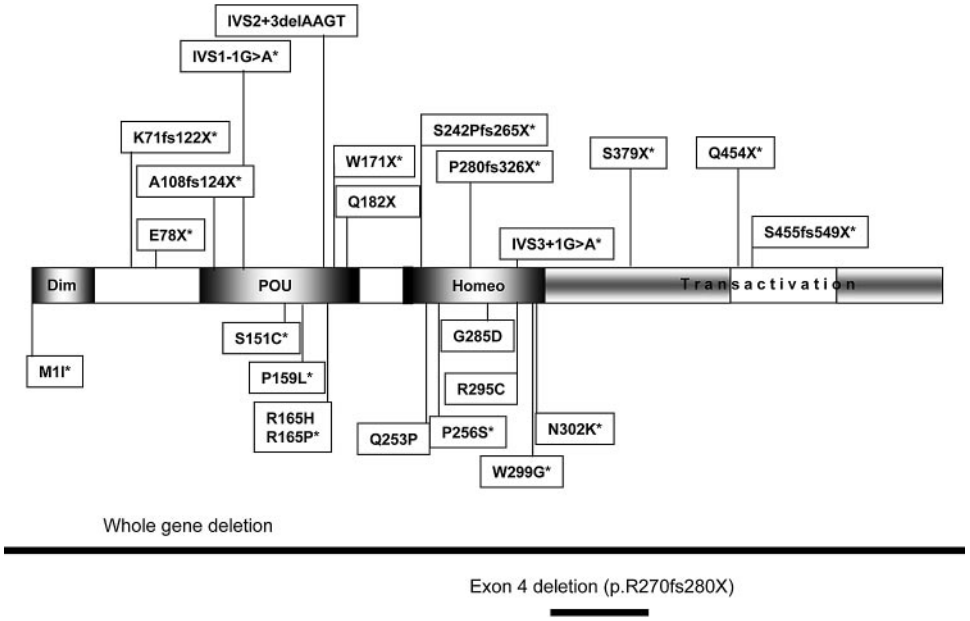

Figure 1. HNF1β protein and localization of the various mutations identified in this study. The N-terminal portion of the protein consists of a short dimerization domain (dim). The DNA-binding domain is characterized by a region distantly related to the POU box-specific domain and an atypical homeodomain structure. The residues required for HNF1β transactivation have been mapped to the carboxy-terminal region. Deletions are indicated by a solid line. *Novel mutation.

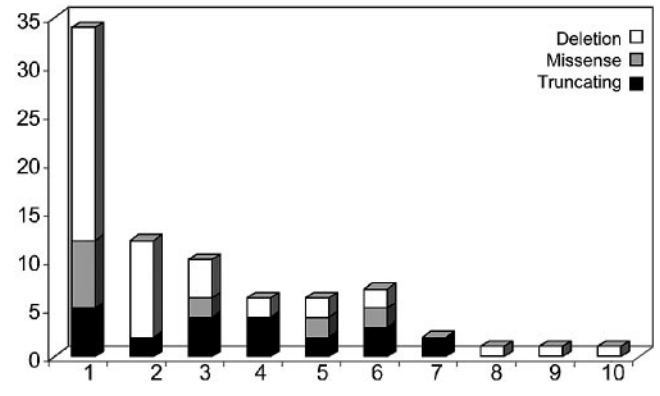

Figure 2. Type of *HNF1B* mutation (□, deletion of the entire gene; ▨, missense mutations; ■, truncating mutations) according to the renal phenotype in patients and affected relatives. 1, prenatal hyperechogenic kidneys; 2, hyperechogenic kidney diagnosed after birth; 3, MCD; 4, unilateral renal agenesis; 5, cystic disease; 6, renal hypoplasia; 7, tubulointerstitial nephritis; 8, pyeloureteral junction; 9, pelvic kidney; 10, lack of renal anomaly.

isolated bilateral hyperechogenic fetal kidneys with normal or slightly enlarged ($\leq$+3 SD) size were the most frequent phenotype observed before birth in patients who carried an *HNF1B* mutation (3); however, one limit of our study is that our population represents patients who had congenital anomalies of the kidney and urinary tract and whose samples were received for *HNF1B* testing in France during a certain period of time. Thus, it will be of interest to perform a prospective study that includes all hyperechogenic kidneys with normal or slightly enlarged size diagnosed before birth and test them for *HNF1B* mutation. Almost all patients with *HNF1B* mutation and moderately enlarged hyperechogenic kidneys before birth displayed normal-sized or small kidneys with hyperechogenicity and/or cortical cysts in the postnatal period, suggesting a slow-down in kidney growth after birth.

Besides hyperechogenic kidneys, *HNF1B* mutations were associated with several other prenatal renal abnormalities but far less frequently: bilateral or unilateral MCD, unilateral renal agenesis, kidney hypoplasia, isolated pyelic dilation, or kidneys with individualized cysts. Because unilateral renal agenesis has been reported in association with *HNF1B* abnormalities only in adults so far (5), it had been suggested that these cases may be due to involution of overlooked MCD (13). Our study shows that genuine renal unilateral agenesis can be associated with *HNF1B* mutation. The absence of cases of bilateral agenesis may be due to the small number of patients tested. In all cases of renal unilateral agenesis associated with *HNF1B* mutation, the single kidney was abnormal. More generally, except for one patient with unilateral hypoplasia and normal contralateral kidney, all probands who carried *HNF1B* mutation displayed bilateral kidney abnormalities. Regarding extrarenal symptoms, no patient with *HNF1B* mutation developed diabetes during early childhood. Only four presented diabetes at 17, 20, 31, and 42 years, respectively, and one developed gestational diabetes. Six other probands had family history of diabetes, but the type of diabetes in relatives was not always known.

Twelve patients with *HNF1B* mutation had early gout and/or hyperuricemia, a feature that has been reported in patients with *HNF1B* mutations (15), but this frequency must be underestimated because the uricemia dosage was not available for many patients in our cohort. Only one adult proband who presented with tubulointerstitial nephropathy and early hyperuricemia that was previously shown not to be associated with *UMOD* mutation was carrying an *HNF1B* mutation. The association of familial hyperuricemic nephropathy with *HNF1B* mutation has been reported previously (5,15), but the mechanisms responsible for the reduced fractional excretion of uric acid are not well understood. HNF1α/HNF1β heterodimers have been shown to bind and positively regulate the proximal promoter region of *SLC22A12*, encoding a transporter that is

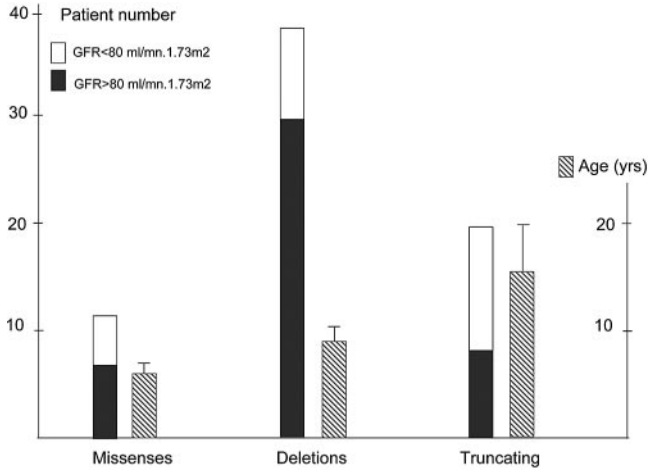

Figure 3. Type of *HNF1B* mutation (deletion, missense mutations, truncating mutations) according to the GFR at last follow-up (■, GFR >80 ml/min per 1.73 m^2; □, GFR <80 ml/min per 1.73 m^2) and age at last follow-up (▨).

responsible for the resorption of urate in the apical membrane of the renal proximal tubule (16); therefore, loss of function would be expected to lead to hypouricemia. The overlap between phenotypes associated with *HNF1B* loss of function and familial *UMOD* hyperuricemic nephropathy may not seem surprising, because *UMOD* was shown to be a target of HNF1β (10); however, familial hyperuricemic nephropathy associated with *UMOD* mutations is thought to be due to a defect in uromodulin transport, associated with a dominant effect, rather than to haploinsufficiency. Thus, the development of the same phenotype associated with *HNF1B* haploinsufficiency is not fully understood. Nevertheless, the finding of hyperuricemia and/or of low uric acid excretion fraction should be an additional argument to screen for *HNF1B* mutation in patients who present with congenital anomalies of the kidney and urinary tract.

Hypomagnesemia was also found in several individuals with *HNF1B* mutation, although blood magnesium dosage was not always performed. Low plasma magnesium level was recently reported by another group and may be related to the transcriptional regulation of *FXYD2* by HNF1β (6). In addition to the frequent and moderate elevation of liver enzymes that was previously reported (17), we observed a severe cholestasis with pruritus in the affected mother of one patient. Cholestasis associated with *HNF1B* mutation was previously reported (18) and is not unexpected given the known role of HNF1β in bile duct morphogenesis (19).

Both the type and the severity of the renal disease were variable in this series, and our data show that *HNF1B* mutations can be associated with very severe prenatal renal failure (in four probands, the pregnancy was terminated because of anamnios, and termination of previous pregnancy for severe renal disease with anamnios was reported in relatives in two additional families) as well as with normal renal function in adulthood. In our series, as in others, there was no obvious correlation between the type of mutation and the type and/or severity of renal disease. We observed both inter- and intrafamilial variability of the phenotype in patients who harbored the same mutation. The lack of genotype–phenotype correlation and the wide variability observed within a given family make the genetic counseling particularly difficult in these families.

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Disclosures

None.

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