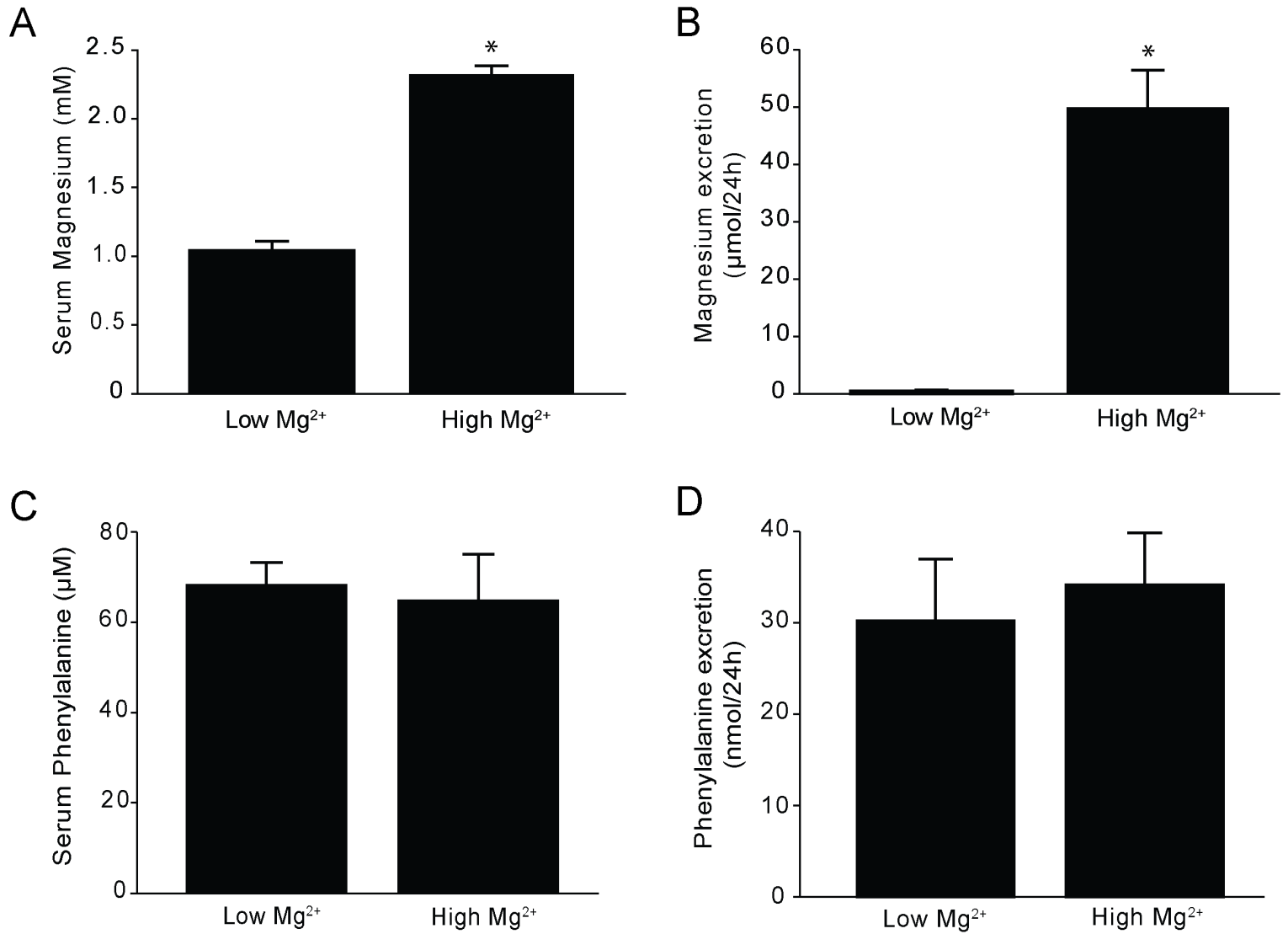


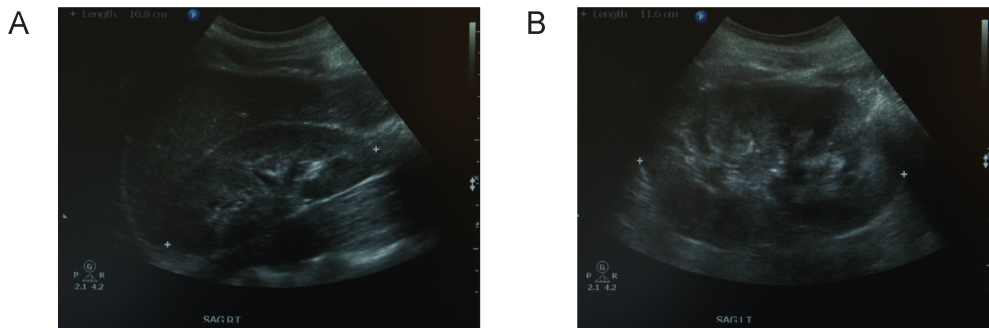
Supplemental Figure 1



Supplemental Figure 1. Effect of dietary Mg^{2+} on serum and urinary Mg^{2+} and phenylalanine levels. C57Bl/6 mice were fed low (0.02% wt/wt) or high (0.48% wt/wt) Mg^{2+} -containing diets for 15 days. Before sacrifice, serum and 24h-urines samples were collected for Mg^{2+} (A and B) and phenylalanine (C and D) levels determination. Results are depicted as mean \pm SEM. *, $p < 0.05$ versus low Mg^{2+} (n=10).

Supplemental Figure 2

Patient 1 (BIODEF 272)

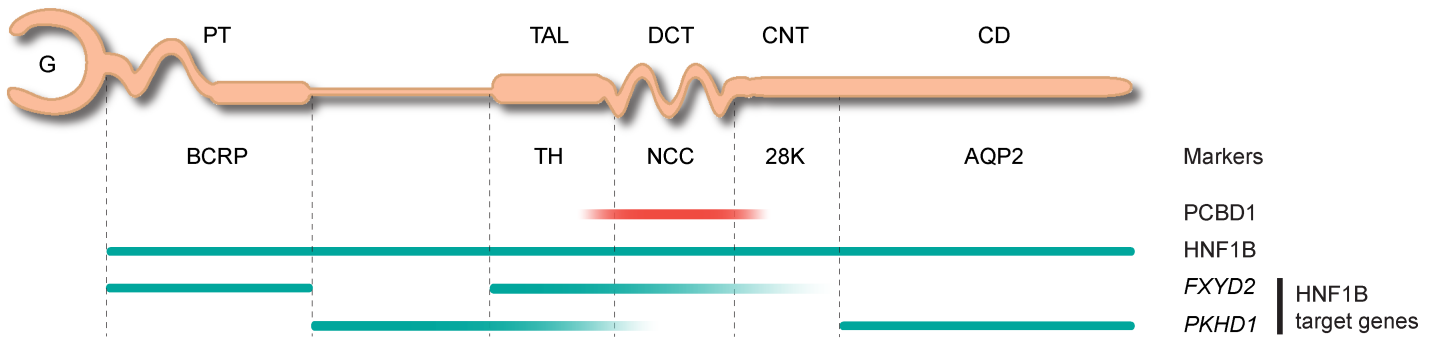


Patient 2 (BIODEF 329)



Supplemental Figure 2. Abdominal ultrasound examinations of patient 1 and 2. In patient 1, size of the right (A) and left (B) kidney was normal, with no cysts. Ultrasound analysis of kidney (C), pancreas (D) and liver (E) in patient 2 showed normal organ morphology.

Supplemental Figure 3



Supplemental Figure 3. Schematic diagram of the expression pattern of PCBD1, HNF1B and two relevant HNF1B target genes along the nephron. Immunohistochemical analysis revealed that the PCBD1 protein colocalizes with NCC expression in DCT and in part with TH and 28K expression in TAL and CNT, respectively. Since HNF1B is expressed in all epithelial cells of the renal tubules, PCBD1 will enhance the transcription of HNF1B target genes that are expressed in DCT, e.g. *FXYD2*. The transcription of HNF1B target genes in nephron segments rather than DCT is not influenced by PCBD1. Therefore, mutations in *PCBD1* are likely to be causative for a renal disease resembling the phenotype observed in patients with *FXYD2*, but not *PKHD1*, defects. G: glomerulus; PT: proximal tubule; TAL: thick ascending loop of Henle; DCT: distal convoluted tubule; CNT: connecting tubule; CD: collecting duct; BCRP, breast cancer resistance protein; TH, Tamm Horsfall; NCC, Na⁺/Cl⁻ cotransporter; 28K, calbindin-D_{28K}; AQP2, aquaporin-2; *FXYD2*, gene encoding for the γ -subunit of the Na⁺/K⁺ ATPase, isoform α ; *PKHD1*, gene encoding for fibrocystin.

Supplemental Table 1. Primer sequences used for real-time PCR analysis.

Gene product	Forward (5'-3')	Reverse (5'-3')
<i>Pcbd1</i>	TGGACATGGCCGGCAAGGC	CCCACAGCCCTCAGGTTTG
<i>Hnf1b</i>	CAAGATGTCAGGAGTGCGCTAC	CTGGTCACCATGGCACTGTTAC
<i>Gapdh</i>	TAACATCAAATGGGGTGAGG	GGTTCACACCCATCACAAC

Pcbd1: pterin-4 alpha-carbinolamine dehydratase/dimerization cofactor of hepatocyte nuclear factor 1 alpha; *Hnf1b*: hepatocyte nuclear factor b; *Gapdh*: glyceraldehyde 3-phosphate dehydrogenase.

Supplemental Table 2. Primer sequences used for cloning or mutagenesis PCR

Gene product	Primer sequences (5'-3')	
HA-PCBD1 wild-type	F	CGACCGGTGGCTGGCAAGCACACAGG
	R	CGCTCGAGCTATGTCATGGACACTGCTAC
Flag-PCBD1 wild-type	F	GCGCTAGCGCCACCATGGACTACAAGGATGACAAGGCTGGCAAAGCACACAGGCTG
	R	CGCTCGAGCTATGTCATGGACACTGCTAC
HA-PCBD1 Glu26Ter	F	GCTGTGGGGTGAATTAGCTGGAAGGCCGTGAT
	R	ATCACGGCCTTCCAGCTAATTCCACCCACAGC
HA-PCBD1 Thr78Ile	F	CACATCACGCTGAGCATCCATGAGTGTGCCGGC
	R	GCCGGCACACTCATGGATGCTCAGCGTGATGTG
HA-PCBD1 Cys81Arg	F	AGCACCCATGAGCGTGCCGGCCTTTCA
	R	TGAAAGGCCGGCACGCTCATGGGTGCT
HA-PCBD1 Glu86Ter	F	TGTGCCGGCCTTTCATAACGGGACATAAACCTG
	R	CAGGTTTATGTCCCGTTATGAAAGGCCGGCACA
HA-PCBD1 Arg87Gln	F	GCCGGCCTTTCAGAACAGGACATAAACCTGGCC
	R	GGCCAGGTTTATGTCCTGTTCTGAAAGGCCGGC
HA-PCBD1 Glu96Lys	F	CTGGCCAGCTTCATCAAACAAGTAGCAGTGTC
	R	GGACACTGCTACTTGTGGATGAAGCTGGCCAG
HA-PCBD1 Glu97Ter	F	GCCAGCTTCATCGAATAAGTAGCAGTGCCATG
	R	CATGGACACTGCTACTTATTTCGATGAAGCTGGC
Flag-HNF1B wild-type	F	CGGCTAGCCCACCATGGACTACAAGGATGACGATGACAAGTGCGCGCCCGTGTCCAAG
	R	GGCCCCCATTGTAACCGGTCG
HA-HNF1B wild-type	F	GGCGCGCCATGGTGTCCAAGCTCACGTCGC
	R	TCTAGATCACCAGGCTTGTAGAGGACAC
HA-HNF1B Δ2-30	F	CGGGCGCGCCGAGGAGTTGCTGCCATC
	R	GGCCCCCATTGTAACCGGTCG
HA-HNF1B Lys156Glu	F	CCCAGCATCTCAACGAGGGCACCCCTATG
	R	CATAGGGGTGCCCTCGTTGAGATGCTGGG
HA-HNF1B Gln253Pro	F	CAGGCCTACGATCGGCCAAAGAACCCAGCAAG
	R	CTTGCTGGGGTTCTTTGGCCGATCGTAGGCCTG
HA-HNF1B Arg276Gly	F	GCAGAATGTTTGCAGGGAGGGGTGTCCCCCTC
	R	GAGGGGGACACCCCTCcCTGCAAACATTCTGC
HA-HNF1B His324Ser325fsdelCA	F	CTCCAACCAGACTCAGCCTGAACCCTCTGC
	R	GCAGAGGGTTCAGGCTGAGTCTGGTTGGAG
HA-HNF1B Tyr352fsinsA	F	GTCAGGAGTGCCTAACAGCCAGCAGGGAAAC
	R	GTTTCCCTGCTGGCTGTTAGCGCACTCCTGAC
PKHD1 exon 1/-1.5 kb	F	CGGGGTACCGAAAATTGGGGTGATTTTAGGATG
	R	GGGAAGCTTCTTTTTTTCTGTTTCTGTCTCC

PCBD1: pterin-4 alpha-carbinolamine dehydratase/dimerization cofactor of hepatocyte nuclear factor 1 alpha; HNF1B: hepatocyte nuclear factor b; PKHD1: polycystic kidney and hepatic disease 1.