Supplemental Table 1: **Human and mouse PC1 sequence equivalencies**

Human	Mouse	Domain	Clinical significance; Score*	PolyPhen prediction; PSIC score difference
C210G	C210G	WSC	Highly likely pathogenic; 15	Probably damaging; 3.410
V690D	V685D	Between C-Lectin & PKD-2	Highly likely pathogenic; 13	Probably damaging; 2.012
G1166S	G1160S	PKD-5	Likely pathogenic; 10	Possibly damaging; 1.824
R2089	C2085R	PKD-15	No report	Possibly damaging; 1.756
L2816P	L2811P	REJ	Likely pathogenic; 6	Probably damaging; 2.040
L3048H	L3040H	GPS	No report	Probably damaging; 2.243
Y4109X	Y4100X	COOH-terminus	No report	ND
R4213X	R4204X	COOH-terminus	No report	ND
R4228X	R4218X	COOH-terminus	Definitely pathogenic	ND

*From: http://pkdb.mayo.edu/cgi-bin/mutations.cgi:
Highly likely pathogenic: $\geq$11
Likely pathogenic: 5-10
Indeterminate: $\leq$4
Definitely pathogenic: nonsense mutation

Supplemental Table 2: **Genes contained in BAC RPCI22-287A3**

Gene Symbol	Gene name
Traf7**	TNF receptor-associated factor 7
Rab26	RAB26, member RAS oncogene family
Pkd1	**Polycystic kidney disease gene 1**
Tsc2	tuberous sclerosis 2
Nthl 1	Nth endonuclease III-like 1
Slc9a3r2	Solute carrier family 9 (sodium/hydron exchanger), member 3 regulator
Npw	Neuropeptide W
Zfp598	Zinc finger protein 598
Syngr3	Synaptogrin3
Gfer**	Growth factor, augmenter of liver regeneration

*Genes are list in order from the centromeric→telomeric orientation on the chromosome.
**Partial genes.

Supplemental Table 3: **Expression levels of PC1 in *Pkd1$^{F/H}$-BAC* transgenic mouse lines.**

Transgenic line #	Tg248	Tg276	Tg8	Tg14
qPCR copy number	2	3	8	1
Relative protein expression*	3	3.5	2.1	1.0
Functional rescue	yes	yes	yes	yes

* Relative protein expression is normalized to relative expression from transgenic line #14.
ND, not determine.

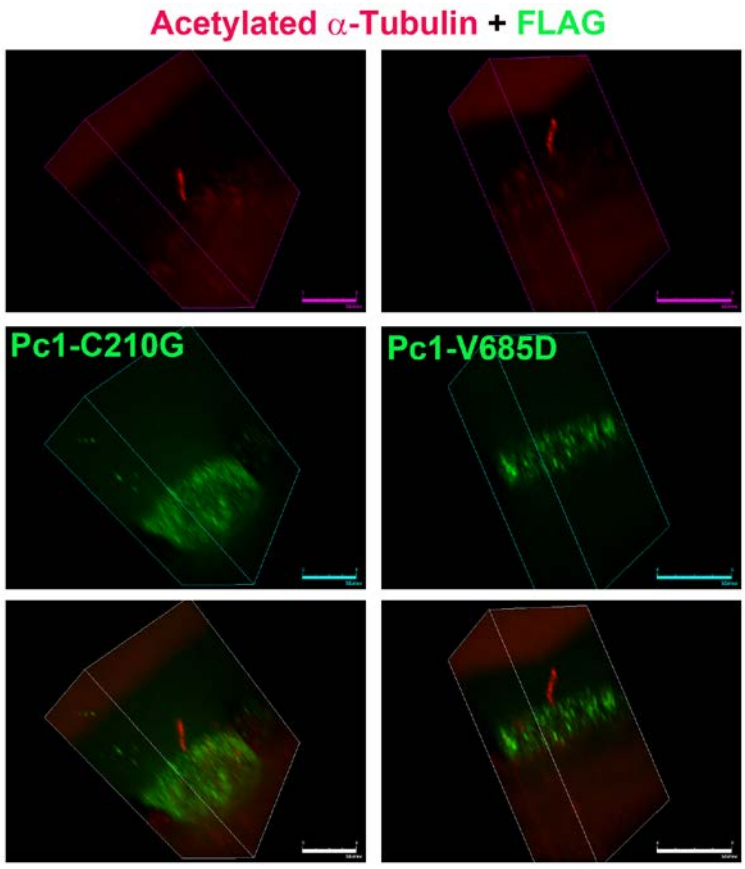
Supplemental Table 4: Functional rescue of embryonic lethality of *Pkd1*$^{-/-}$ mice by the *Pkd1*$^{F/H}$*-BAC* transgenic lines.

Transgenic line #	*Pkd1*$^{+/+}$;*Pkd1*$^{F/H}$*-BAC*	*Pkd1*$^{+/-}$;*Pkd1*$^{F/H}$*-BAC*	*Pkd1*$^{-/-}$;*Pkd1*$^{F/H}$*-BAC*
Tg248	2 (22%)	3 (33%)	4 (45%)
Tg276	8 (20%)	24 (60%)	8 (20%)
Tg8	7 (35%)	7 (35%)	6 (30%)
Tg14	5 (28%)	9 (50%)	4 (22%)
Totals	37 (27%)	66 (48%)	34 (25%)

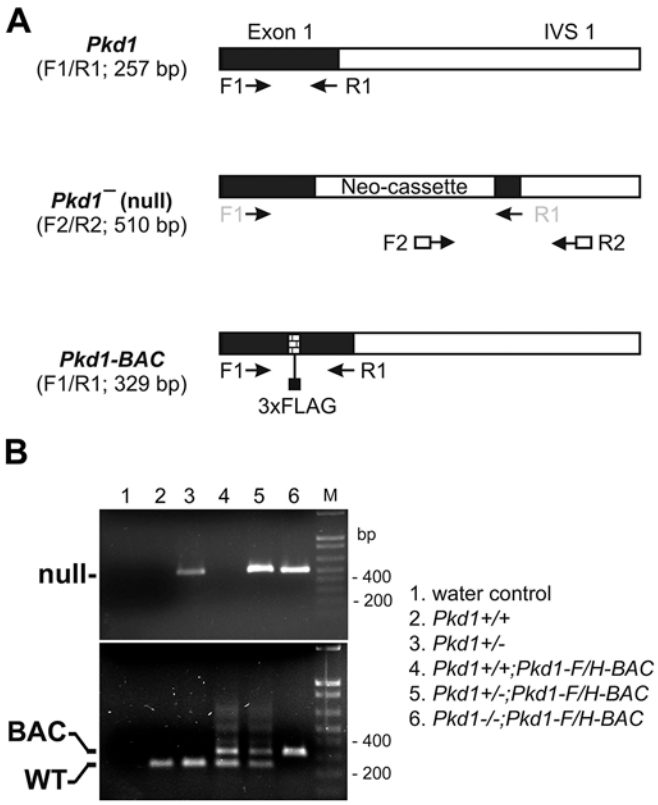
Supplemental Table 5: Failure of functional rescue of embryonic lethality of *Pkd1*$^{-/-}$ mice by the *Pkd1*^{L3040H}*-BAC* transgenic lines.

Transgenic line #	qPCR copy number	*Pkd1*$^{+/+}$;*Pkd1*^{L3040H}	*Pkd1*$^{+/-}$;*Pkd1*^{L3040H}	*Pkd1*$^{-/-}$;*Pkd1*^{L3040H}
Tg46	1	13 (25%)	39 (75%)	0 (0%)
Tg7	3	9 (41%)	13 (59%)	0 (0%)
Tg9	1	5 (33%)	10 (67%)	0 (0%)
Totals	N/A	27 (30%)	62 (70%)	0 (0%)

N/A: not applicable.

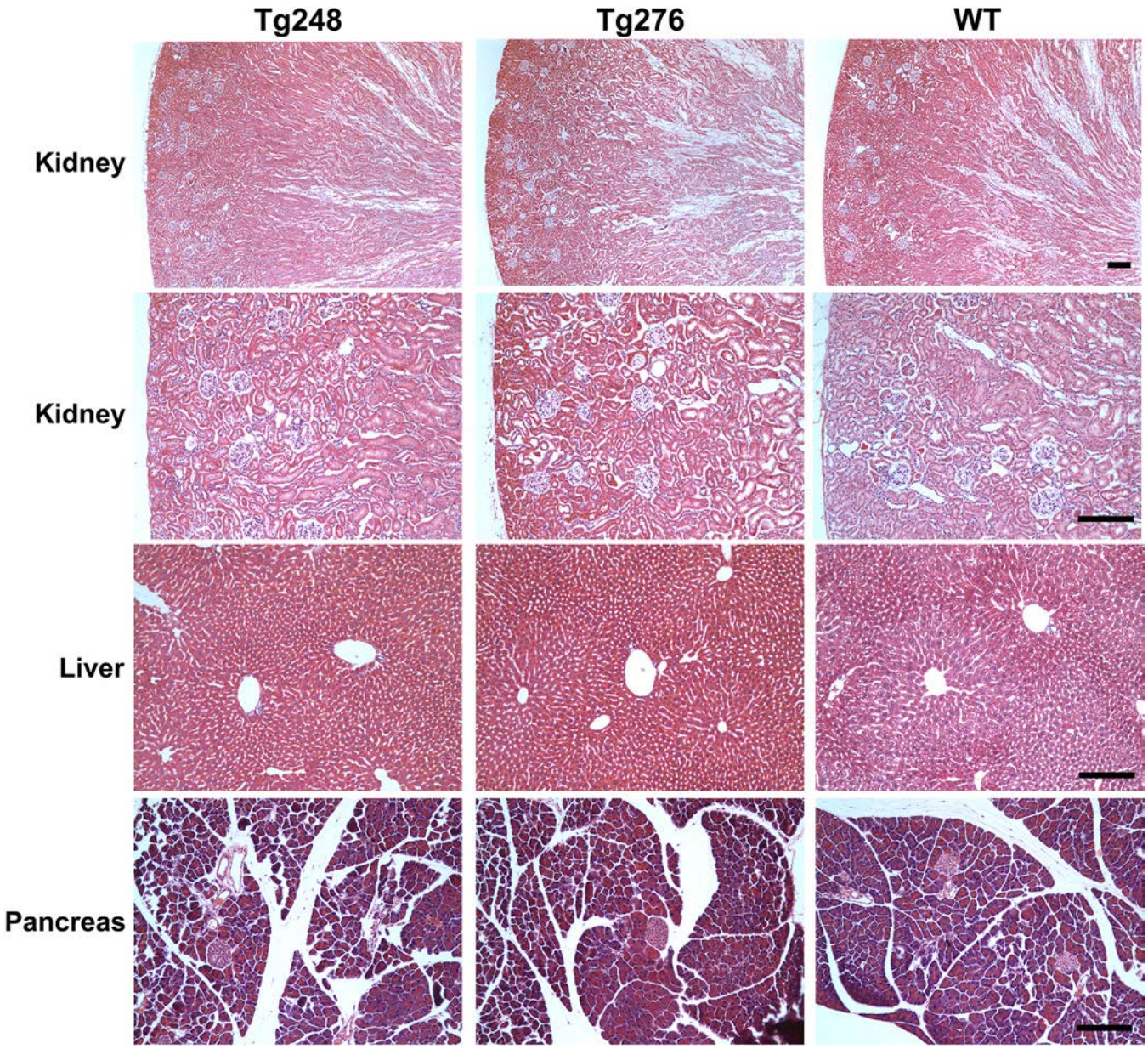


Supplemental Figure 1
Alternate projections from a 3D view assembled from z-stack images showing abundant expression of the respective Pc1 pathogenic missense mutations, Pc1-C210G and Pc1-V685G, in the cell bodies (green) and absence from cilia (red). These are the same mutant variants presented in Fig. 3D and 3E, respectively. Scale bar, 8 μm.



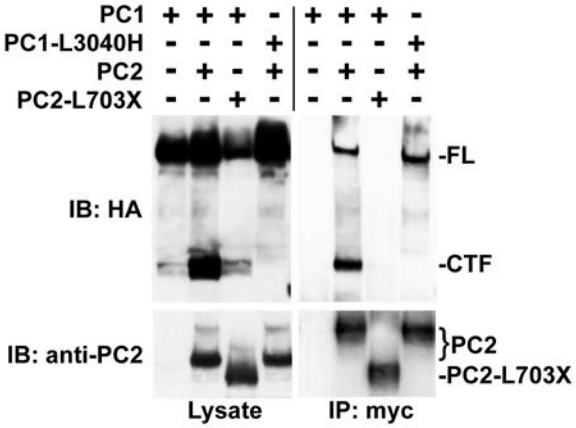
Supplemental Figure 2

Strategy of the PCR genotyping endogenous and BAC alleles of *Pkd1*. (**A**) Schematic representation of the WT (*Pkd1*), null (*Pkd1*$^{-}$) and FLAG-tagged BAC (*Pkd1*$^{F/H}$*-BAC*) alleles showing location of genotyping primers and respective product sizes. (**B**) Representative PCR genotyping for mice with the indicated genotypes.



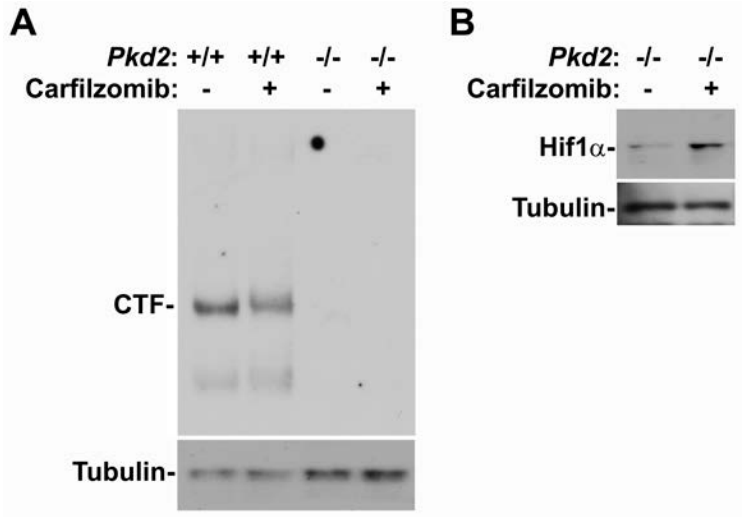
Supplemental Figure 3

Functional rescue of *Pkd1*$^{-/-}$ mice by the *Pkd1*$^{F/H}$*-BAC* transgene. Kidney, liver and pancreas tissue sections stained with hematoxylin and eosin from 6 month old mice with genotypes *Pkd1*$^{-/-}$;*Pkd1*$^{F/H}$*-BAC (Tg248)*, *Pkd1*$^{-/-}$;*Pkd1*$^{F/H}$*-BAC (Tg276)*, and wild type (WT). The respective *Pkd1*$^{F/H}$*-BAC* transgenes completely rescue the *Pkd1*$^{-/-}$ phenotype. Size bars, 500 μm.

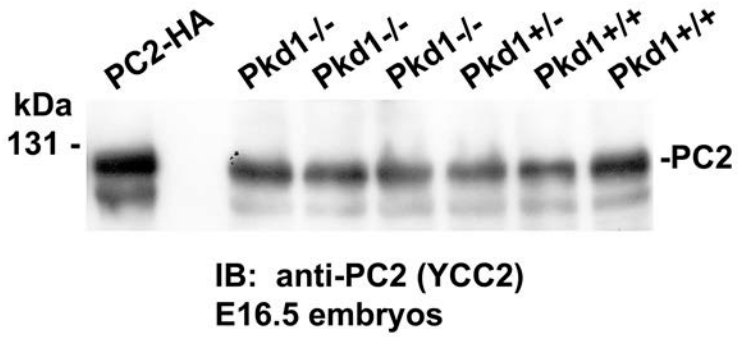


Supplemental Figure 4

GPS cleavage-deficient Pc1^{L3040H} interacts with PC2. PC1^{L3040H} does not undergo GPS cleavage (*top left panel*) but interacts with PC2 as indicated by co-immunoprecipitation (*right panels*). COOH-terminal truncated PC2^{L703X} does not interact with PC1 and does not co-IP with it. Co-expression of wild type PC2 increases expression of PC1-CTF (*top left panel*), whereas co-expression with PC2L^{703X} does not. PC2-myc, PC2^{L703X}-myc, PC1 and PC1^{L3040H} were transiently expressed in COS7 cells as indicated. Lysates were IP using anti-myc. PC1 and PC1^{L3040H} were detected by immunoblotting with anti-HA antibodies while PC2 and PC2^{L703X} were detected using YCB9 anti-PC2 antisera.



Supplemental Figure 5
Proteasomal Inhibition does not restore Pc1-CTF levels in the absence of Pc2. (**A**) SF4 *Pkd2*$^{fl/fl}$*;Pkd1*$^{F/H}$*-BAC* cells (+/+) and SF3A *Pkd2*$^{-/-}$*;Pkd1*$^{F/H}$*-BAC* cells (–/–) with and without treatment with carfilzomib (100 nM for 16 hours) followed by immunoblotting with anti-HA to detect Pc1-CTF. Carfilzomib treatment did not increase in the steady state level of Pc1-CTF. (**B**) Effective inhibition of proteasome function in SF3A cells was documented by increased Hif1α expression in carfilzomib-treated cells compared with DMSO treated controls. Tubulin serves as loading control.



Supplemental Figure 6

Loss of Pc1 does not affect steady state levels of Pc2. Whole tissue lysates of E16.5 mouse embryos with the indicated genotypes show no effect on Pc2 expression. PC2-HA is transfected cell lysate.