

Supplementary Table 1. Means and standard deviations of phenotypic measurement of $PRCP^{gt/gt}$ and congenic strains B6.C-D7Mit353 and B6.C-D7Mit373. The sample size for each group is indicated in the column header ($PRCP^{gt/gt}$: PRCP gene-trap; $PRCP^{wt/wt}$: wildtype strain; C353: B6.C-D7Mit353 congenic strain; C373: B6.C-D7Mit373 congenic strain; B353 and B373: C57BL/6ByJ background strains). All mice were male.

Phenotype	PRCPwt/wt (count=14)	PRCPgt/gt (count=19)	B353 (count =49)	C353 (count = 32)	B373 C373 (count =25)	(count = 28)
Body Weight	29.6±1.0	26.6 ±0.5	26.9 ±3.2	27.0 ±4.1	28.5 ±2.5	27.6 ±2.4
Body Length	9.84±0.08	9.37±0.09	9.85±0.43	9.88±0.49	10.1 ± 063	9.94 ± 070
Retroperitoneal WAT	0.28 ±0.012	0.17 ±0.009	0.39 ±0.17	0.3 ±0.17	0.35 ±0.16	0.27 ±0.16
Femoral WAT	0.11 ±0.008	0.05 ±0.004	0.7 ±0.3	0.55 ±0.33	0.72 ±0.32	0.56 ±0.3
Gonadal WAT	0.19 ±0.009	0.07 ±0.007	0.24 ±0.12	0.2 ±0.13	0.23 ±0.11	0.17 ±0.12
Mesenteric WAT	0.59 ±0.03	0.33 ±0.03	0.27 ±0.15	0.23 ±0.21	0.27 ±0.14	0.19 ±0.12
Body Mass Index	3.005 ±0.095	2.84 ±0.047	2.738 ±0.03	2.73 ±0.05	2.82 ±0.03	2.78 ±0.03
Total Fat	1.17 ±0.062	0.62 ±0.045	1.6 ±0.7	1.3 ±0.8	1.6 ±0.7	1.2 ±0.7
Adiposity Index	0.039 ±0.001	0.023 ±0.0014	0.059 ±0.022	0.47±0.028	0.055 ±0.021	0.043±0.02

Supplementary Table 2. SNPs that define the borders of the 373 congenic donor region.

SNPs are denoted by their base pair position in the coding region (Pcf11 & Odz4) or gene fragment transcript position (NM_021427.1). SNPs are shown as changes from the C57BL/6J base to the BALB/c base. In NM_021427.1, deletions occur in BALB/c sequence.

Gene Name	Ensembl Accession	SNPs	Coding Changes
Pcf11	ENSMUSG000000041328	T2916C	Q972Q
Pcf11	ENSMUSG000000041328	G4224T	T1408T
NM_021427.1	ENSMUSG000000051515	A1319T	N/A
NM_021427.1	ENSMUSG000000051515	del 1395-1397	N/A
NM_021427.1	ENSMUSG000000051515	del 1401	N/A
Odz4	ENSMUSG000000048078	A7843G	K2615E
Odz4	ENSMUSG000000048078	G8137A	V2713M

del=deletion

Supplementary Table 3. GenBank accession numbers for gene sequences containing our SNPs.

Gene	Strain	GenBank Accession
Pcf11	BALB/c	EF537860
Pcf11	C57BL/6J	EF537861
NM_021427.1	BALB/c	EF537862
NM_021427.1	C57BL/6J	EF537863
Odz4	BALB/c	EF537864
Odz4	C57BL/6J	EF537865

Supplementary Table 4. Sequencing primers for Prcp, quantitative PCR primers for Prcp and sequencing primers for 373 subcongenic border genes.

Gene Name	Experiment	Primer Sequence
Prcp	sequencing of Prcp	1upstreamprox: 5'-CAAAGGACACAGACAAGCCC-3'
Prcp	sequencing of Prcp	1upstreamproxr: 5'-CCACACTCAACCAGAACAGC-3'
Prcp	sequencing of Prcp	2upstreamprox: 5'-GTCCAACCTCCGTCTGGAAGG-3'
Prcp	sequencing of Prcp	2upstreamproxr: 5'-AAGACAGGAGCAGGAGAGCC-3'
Prcp	sequencing of Prcp	5'UTRprox: 5'-TGCCTGGTTTTAGGTTTCCC-3'
Prcp	sequencing of Prcp	5'UTRproxr: 5'-AAGACAGGAGCAGGAGAGCC-3'
Prcp	sequencing of Prcp	1prox: 5'-CTTTAGTCGCCCACACTGTG-3'
Prcp	sequencing of Prcp	1proxr: 5'-CAGGTTACCCAGGTCTCAG-3'
Prcp	sequencing of Prcp	2prox: 5'-CAGCCTGGTTCAGGATGAAG-3'
Prcp	sequencing of Prcp	2proxr: 5'-CACAGAGGAAGGATCGAAGG-3'
Prcp	sequencing of Prcp	3prox: 5'-CTATCAACATCCATGACGGGGC-3'
Prcp	sequencing of Prcp	3proxr: 5'-TTGGCAGAATGTAGCCAGAC-3'
Prcp	sequencing of Prcp	3'UTRprox: 5'-GCCTTGTCTGCTCTCTTTG-3'
Prcp	sequencing of Prcp	3'UTRproxr: 5'-AAGTCCCCACACACCTGATG-3'
Prcp	quantitative PCR	prcpabif: 5'-GAACTACCCTTACGCATGCAACT-3'
Prcp	quantitative PCR	prcpabir: 5'-AATATTGGCACACCTCCTTGATG-3'
Abrp	quantitative PCR	AbrpF: 5'-GGACCCGAGAAGACCTCCTCCTT-3'
Abrp	quantitative PCR	AbrpR: 5'-TCAATGGTGCCTCTGGAGATT-3'
Pcf11	sequencing 373 borders	1prox: 5'-CAAGATTTGCTGGCCTTGAT-3'

Pcf11	sequencing 373 borders	1proxr: 5'-GACCCTCAAACCTCATTCCA-3'
Pcf11	sequencing 373 borders	2proxf: 5'-GTTTGGCCAAGGACAACAGT-3'
Pcf11	sequencing 373 borders	2proxr: 5'-TGCCATTCTTCCTCCTCTTC-3'
NM_021427.1	sequencing 373 borders	1proxf: 5'-CGATGGTGGGGAACCTACTA-3'
NM_021427.1	sequencing 373 borders	1proxr: 5'-CTTGATCATCCCACCCAAAT-3'
NM_021427.1	sequencing 373 borders	2proxf: 5'-ACTGACTGCGAGGAACTCGT-3'
NM_021427.1	sequencing 373 borders	2proxr: 5'-CAAAAACCAAGGAAGGCAAA-3'
Odz4	sequencing 373 borders	1proxf: 5'-GGGTCAAGTTTGCCTTGAAA-3'
Odz4	sequencing 373 borders	1proxr: 5'-AGTCAGGAATACGGCCACAG-3'

Supplementary Table 5. PRCP quantitative real time PCR in brain tissues.

Ct means and standard deviations with one Syber Green probe and 3 TaqMan probes covering different exons. Endogenous controls were used to normalize data: Beta-2 microglobulin (B2m), TATA binding protein (Tbp), Glyceraldehyde-3-phosphate dehydrogenase (Gapdh) and Beta-glucuronidase (Gus) for the ABI TaqMan probes. Fold changes (FC) were calculated as the average 7 or 8 biological replicates. P-values were calculated using one-tailed Student t-tests with 7 or 8 samples per group. **Background:** C57BL/6ByJ strain; **Heterozygote:** B6.CD7Mit353 heterozygote congenic strain; **Homozygote:** B6.C-D7Mit353 homozygote congenic strain.

	Syber Green			ABI Taqman									Prcp qPCR	
Congenic	PRCP exon 5			Probe Exon 3-4			Probe exon 6-7			Probe Exon 2-3			Average	
Genotype	Ct ±SD	FC *	p-value	Ct ±SD	FC *	p-value	Ct ±SD	FC *	p-value	Ct ±SD	FC *	p-value	FC *	SD
Background	27.8 ±0.4	1	-	30.1 ±0.5	1	-	29.5 ±0.4	1	-	30.8 ±0.3	1	-	1	-
Heterozygote	28.7 ±0.4	0.8	4.E-04	30.8 ±0.4	0.9	5.E-03	30.3 ±0.4	0.8	4.E-04	31.5 ±0.4	0.9	7.E-04	0.8	0.04
Homozygote	28.8 ±0.5	0.5	5.E-04	31.0 ±0.6	0.6	6.E-03	30.3 ±0.6	0.6	6.E-03	32.2 ±0.5	0.4	5.E-06	0.5	0.11

* FC: Fold Change versus the background strain; pvalue calculated using one tailed Student ttest

Supplementary Table 6. Primers for the Characterization of the PRCP^{gt/gt} Mice

Target	Sense Primer	Antisense Primer	Probe
<u>Genotyping*</u>			
β-lac Z	5'-ATGGCCATTACCGTTGATGT-3'	5'-CCAGTTTACCCGCTCTGCTA-3'	5'-56-FAM/GAAGTGGCGAGGGATACAC/3BHQ1/-3'
<u>Insertional Mutation Site*</u>			
E4-TM	5'-GGCATGCTTGCAGCCTGGTT-3'	5'-GTGATCCAGGACTGGGAAGA-3'	5'-56-FAM/TTGTACTTGGGTCCCAGGTC/3BHQ1/-3'
<u>PRCP mRNA</u>			
5'UT-TM	5'-TGCTGCTGCCTCTCTTTCT-3'	5'-GTGATCCAGGACTGGGAAGA-3'	5'-56-FAM/TTGTAGTTGGGTCCCAGGTC/3BHQ1/-3'
5'UT-E8	5'-TGCTGCTGCCTCTCTTTCT-3'	5'-AAAGGGCATGACCATTCTG-3'	5'-56-FAM/TTGTAGTTGGGTCCCAGGTC/3BHQ1/-3'
5'UT-E9	5'-TGCTGCTGCCTCTCTTTCT-3'	5'-CCACGGGTCTAACTCACCAT-3'	5'-56-FAM/TTGTAGTTGGGTCCCAGGTC/3BHQ1/-3'
E4-E8	5'-TTTTGCAGAGTTAATCAGACACTTG-3'	5'-GTCCACAGAAAGGGTTCAA-3'	5'-56-FAM/TTGTAGTTGGGTCCCAGGTC/3BHQ1/-3'

- Genotyping by real time PCR was performed with tail DNA with the indicated primers. The characterization of the insertional mutation site was performed with mRNA from a wild type and PRCP^{gt/gt} mouse.
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- 5'UT stands for the 5' untranslated region; “E” represents the exon region; “TM” stands for the TM region on trapping vector pGT1.1TM