

Fig. S1. PTP1B depletion increases ALDH activity in mammary epithelial cells. (A) Representative flow cytometry dot plots showing the gating strategy used to isolate mammary epithelial cell subpopulations. Cells were first gated on the basis of forward and side scatters (FSC-A and SSC-A), followed by the exclusion of doublets and higher-order cell clumps using a time-of-flight approach (pulse width). Dead cells were then gated out by DAPI staining, followed by the exclusion of leukocytes by CD45 staining. Finally, total epithelial cells were isolated based on their CD24 and Sca1 expression. Further isolation of MaSCs and myoepithelial cells was performed based on the expression of CD24 and CD49f. (B) Mammary repopulating activity of *Ptp1b*^{+/+} and *Ptp1b*^{-/-} MECs. 10,000, 5000, 2000, 1000 and 500 freshly isolated mammary epithelial cells (MECs) from *Ptp1b*^{-/-} and *Ptp1b*^{+/+} glands in estrus were injected into the cleared fat pad of FVB mice. The repopulation of the fat pad was scored 7 weeks after transplantation. The transplantation frequency analysis was performed using R and the statmod package as previously described (Shackleton et al., 2006). (C) (Left) Dot plot pattern of ALDH-positive cells showing an increase in ALDH activity in *Ptp1b*^{-/-} MECs. (Right) The percentages of cells with high ALDH activity (ALDH^{br}) from *Ptp1b*^{+/+} and *Ptp1b*^{-/-} nulliparous mice at estrus. Values represent mean \pm s.e.m. ($n=3$, $*P<0.05$ by Student's *t*-test).

A

Cell cycle genes upregulated in PTP1B -/-					
Accession #	Symbol	description	fold changes	p-value	Adj p-value
NM_009828	CCNA2	cyclin A2	2.46	0.0115	0.1768713
NM_011623	Topo2	topoisomerase (DNA) II alpha	2.30	0.0078	0.1592963
NM_007630	CCNB2	cyclin B2	2.14	0.0144	0.1916421
NM_011121	Plk1	polo-like kinase 1 (Drosophila)	2.14	0.0126	0.1836071
NM_007659	Cdc2a (CDK1)	cell division cycle 2 homolog A (S. pombe)	2.00	0.0104	0.1712105
NM_007691	Chek1	checkpoint kinase 1 homolog (S. pombe)	1.87	0.0244	0.2270902
NM_018754	SFN	stratifin	1.62	0.0005	0.062044
NM_001081117	Mki67	antigen identified by monoclonal Ab Ki 67	1.84	0.0418	0.2758198
NM_025415	CKS2	CDC28 protein kinase regulatory subunit 2	1.48	0.0403	0.272589

estrogen-responsive genes upregulated in PTP1B -/-					
Accession #	Symbol	description	fold changes	p-value	Adj p-value
NM_010118	Egr2	early growth response 2	3.00	2.28E-06	0.0039558
NM_009704	Areg	amphiregulin	2.88	9.55E-09	0.0003318
NM_007969	Expi	extracellular proteinase inhibitor	1.98	0.001761	0.0959992
NM_001081249	Vcan(cspg2)	vesican	1.94	0.001344	0.0889354
NM_010848	c-myb	myeloblastosis oncogene	1.93	3.95E-05	0.0176528
NM_008829	Pgr	Progesterone receptor	1.80	0.005725	0.1446963
NM_010849	c-myc	myelocytomatosis oncogene	1.59	0.002251	0.1048908
NM_011961	Plod2	procollagen lysine, 2-oxoglutarate 5-dioxy	1.57	0.005868	0.1450678
NM_010233	FN1	fibronectin 1	1.51	0.006227	0.1474954
NM_013650	S100A8	S100 calcium binding protein A8 (calgranul	1.46	0.009963	0.1695082
NM_033073	Krt7	keratin 7	1.43	0.007621	0.1586751
NM_019521	Gas6	growth arrest specific 6	1.41	0.002792	0.1120131
NM_008471	Krt19	keratin 19	1.41	0.020774	0.2154201

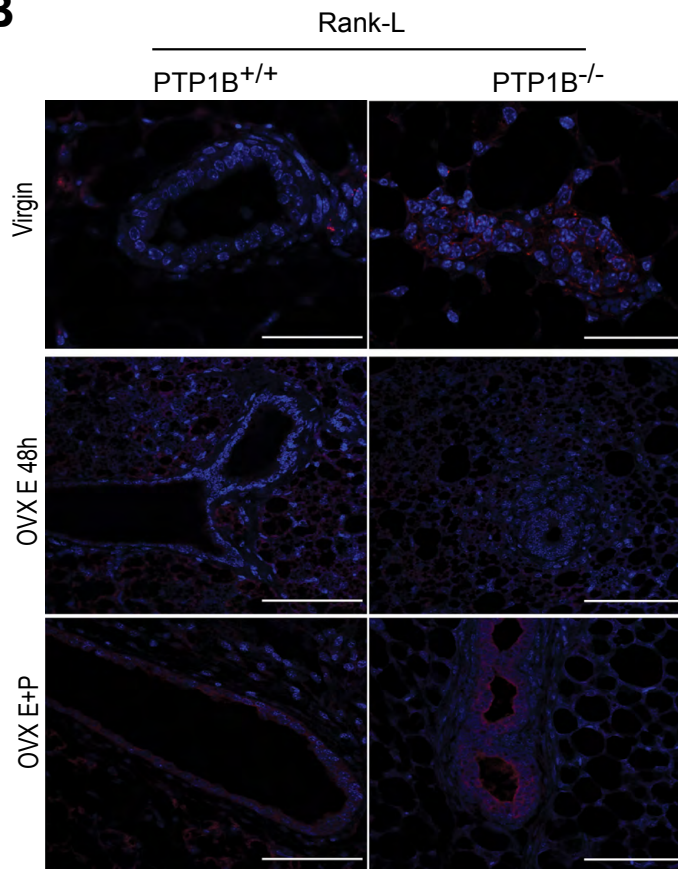
B

Fig. S2. Absence of PTP1B increases expression of cell cycle genes and of the PR target Rankl. (A) Upregulated cell cycle genes (top) and upregulated estrogen-responsive genes (bottom) in nulliparous *Ptp1b*^{-/-} glands compared with wild-type littermates. (B) Representative images of mammary gland sections stained for Rankl. Scale bars: 50 μ m.

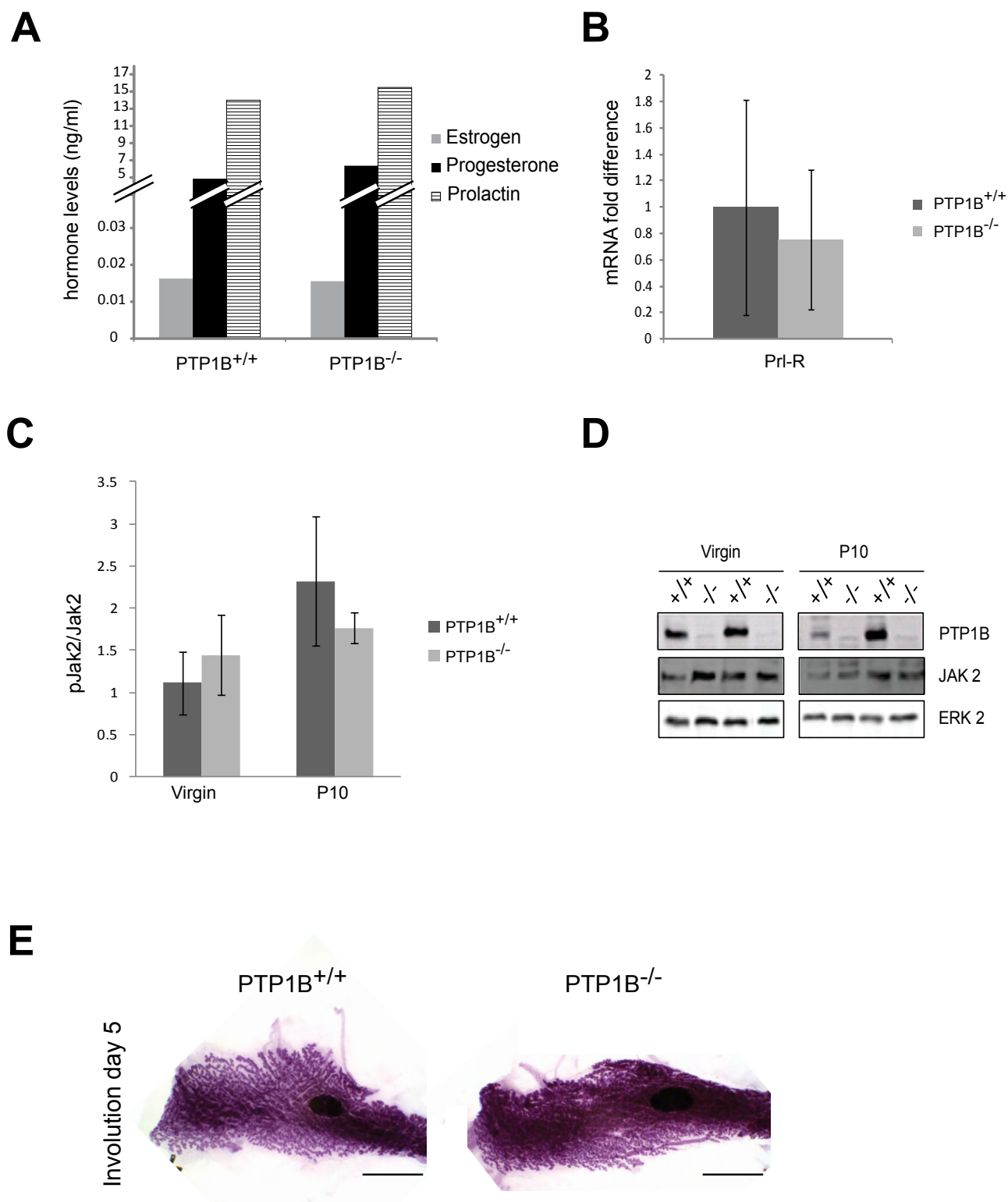


Fig. S3. Absence of PTP1B does not affect plasma levels of pregnancy hormones in mice at estrus and does not alter mammary gland involution. (A) Plasma levels of estrogen, progesterone and prolactin in nulliparous mice at estrus as assessed by ELISA (the plasma of three mice per group was pooled and analyzed using estradiol and progesterone ELISA from DRG Diagnostic and prolactin ELISA from Abcam). (B) Fold changes in Prl-R mRNA as assessed by quantitative real-time PCR. Values represent mean \pm s.e.m. ($n=3$). (C) The ratios of pJak2/Jak2 in virgin and pregnant (day 10) mice. pJak2 levels were assessed using an ELISA assay (Tyr1007/1008, Invitrogen) and normalized to total Jak2 levels assessed by immunoblotting. Values represent mean \pm s.e.m. ($n=3$, $*P<0.05$ by Student's t -test). (D) Immunoblots of lysates from glands in virgin and pregnancy day 10 mice (Jak2 antibody, Cell Signaling). (E) Representative images of whole-mounts of *Ptp1b*^{-/-} and *Ptp1b*^{+/+} mammary glands at involution day 5. Scale bars: 5 mm.

Table S1. Top 19 affected Gene Ontology terms in *Ptp1b*^{-/-} versus *Ptp1b*^{+/+} mammary glands

Gene Ontology term upregulated in <i>Ptp1b</i> ^{-/-}	Cluster frequency	Corrected <i>P</i> -value
Cellular process	244 of 359 genes, 68.0%	4.06E-26
Cell cycle	50 of 359 genes, 13.9%	3.85E-16
Cell division	30 of 359 genes, 8.4%	2.96E-15
Cell cycle phase	35 of 359 genes, 9.7%	4.94E-15
Cell cycle process	40 of 359 genes, 11.1%	7.08E-15
M phase	30 of 359 genes, 8.4%	2.50E-14
Biological regulation	168 of 359 genes, 46.8%	7.51E-14
M phase of mitotic cell cycle	24 of 359 genes, 6.7%	1.55E-12
Cellular component organization or biogenesis	85 of 359 genes, 23.7%	2.32E-12
Cellular component organization	83 of 359 genes, 23.1%	2.34E-12
Nuclear division	23 of 359 genes, 6.4%	6.14E-12
Mitosis	23 of 359 genes, 6.4%	6.14E-12
Mitotic cell cycle	31 of 359 genes, 8.6%	6.45E-12
Organelle fission	23 of 359 genes, 6.4%	1.98E-11
Organelle organization	54 of 359 genes, 15.0%	3.02E-11
Regulation of biological process	157 of 359 genes, 43.7%	3.21E-11
Cellular component organization at cellular level	68 of 359 genes, 18.9%	4.63E-11
Cellular component organization or biogenesis at cellular level	69 of 359 genes, 19.2%	1.29E-10
Metabolic process	159 of 359 genes, 44.3%	9.95E-10

Gene Ontology term downregulated in <i>Ptp1b</i> ^{-/-}	Cluster frequency	Corrected <i>P</i> -value
Alcohol metabolic process	10 of 62 genes, 16.1%	3.16E-06
Small molecule metabolic process	19 of 62 genes, 30.6%	3.18E-06
Glucose metabolic process	7 of 62 genes, 11.3%	5.47E-06
Carbohydrate metabolic process	10 of 62 genes, 16.1%	1.38E-05
Hexose metabolic process	7 of 62 genes, 11.3%	1.88E-05
Cellular carbohydrate metabolic process	9 of 62 genes, 14.5%	2.54E-05
Monosaccharide metabolic process	7 of 62 genes, 11.3%	6.20E-05
Cellular process	44 of 62 genes, 71.0%	0.00011
Carbohydrate transport	5 of 62 genes, 8.1%	0.00059
Skeletal muscle contraction	3 of 62 genes, 4.8%	0.00104
Localization	20 of 62 genes, 32.3%	0.00111
Transport	18 of 62 genes, 29.0%	0.00122
Striated muscle contraction	4 of 62 genes, 6.5%	0.0013
Establishment of localization	18 of 62 genes, 29.0%	0.00157
Biological regulation	33 of 62 genes, 53.2%	0.00245
Regulation of striated muscle contraction	3 of 62 genes, 4.8%	0.0026
Metabolic process	33 of 62 genes, 53.2%	0.0038
Protein homooligomerization	5 of 62 genes, 8.1%	0.00387

List of Gene Ontology terms upregulated and downregulated in *Ptp1b*^{-/-} compared with *Ptp1b*^{+/+} glands from nulliparous mice at estrus. Terms were obtained from the Process Ontology of gene_association.mgi (*P*<0.01).