

SUPPLEMENTARY FIGURE LEGENDS

Supplementary Figure 1. Gab2 deletion ameliorates the excessive proliferation of *Ptpn11*^{E76K/+} myeloid cells in hematopoietic tissues and their infiltration in non-hematopoietic organs. BM, spleens, livers, lungs, and kidneys harvested from *Ptpn11*^{+/+}/*Mx1-Cre*⁺/*Gab2*^{+/+}, *Ptpn11*^{+/+}/*Mx1-Cre*⁺/*Gab2*^{-/-}, *Ptpn11*^{E76K/+}/*Mx1-Cre*⁺/*Gab2*^{+/+}, and *Ptpn11*^{E76K/+}/*Mx1-Cre*⁺/*Gab2*^{-/-} mice were processed for histopathological examination (hematoxylin and eosin staining, 200x).

Supplementary Figure 2. Gab2 depletion does not significantly change the activating effects of the *Ptpn11*^{E76K/+} mutation on HSCs. BM cells freshly harvested from *Ptpn11*^{+/+}/*Mx1-Cre*⁺/*Gab2*^{+/+}, *Ptpn11*^{+/+}/*Mx1-Cre*⁺/*Gab2*^{-/-}, *Ptpn11*^{E76K/+}/*Mx1-Cre*⁺/*Gab2*^{+/+}, and *Ptpn11*^{E76K/+}/*Mx1-Cre*⁺/*Gab2*^{-/-} mice (n=3/group) were analyzed by multiparameter FACS for HSCs, MPP1, MPP2, and MPP3 in the BM. Gating strategies are shown in (A). The percentages (B) and absolute numbers (C) of HSCs, MPP1, MPP2, and MPP3 in the BM were determined. The LSK cell population in the BM were analyzed by multiparameter FACS for apoptotic cells. Gating strategies are shown in (D). Apoptotic cells in the gated LSK cell population were determined (E).

Supplementary Figure 3. Depletion of Gab2 corrects the hypersensitive growth pattern of *Ptpn11*^{E76K/+} myeloid progenitors. BM cells (2x10⁴ cells/mL) freshly harvested from the mice with the indicated genotypes (n=3/group) 8 weeks after pI-pC administration were assessed by CFU assays with the indicated concentrations of IL-3.

Supplementary Figure 4. Depletion of Gab2 decreases cellular responses of *Ptpn11*^{E76K/+} multipotent progenitors to IL-3. LSK (Lineage⁻Sca-1⁺c-Kit⁺) cells were sorted from the mice with the indicated genotypes (n=3/group) and cultured (2x10⁴ cells/well) in IMDM containing 10% FBS and IL-3 (2 ng/mL). After 7 days of *in vitro* culture, the numbers of total cells derived were determined. Three independent experiments were performed with similar results obtained in each. Data shown are mean±S.E.M. from one experiment in triplicates.

Supplementary Figure 5. Rapamycin treatment abrogates hypersensitivity of *Ptpn11*^{E76K/+} myeloid progenitors to IL-3. BM cells were harvested from *Ptpn11*^{+/+}/*Mx1-Cre*⁺ and *Ptpn11*^{E76K/+}/*Mx1-Cre*⁺ mice (n=3/group) after 12-14 weeks of treatment with Rapamycin or vehicle. These cells were assessed by myeloid colony assays with the indicated concentrations of IL-3. Experiments were performed three times with similar results obtained in each. Data shown are mean±S.E.M. from one experiment in triplicates.

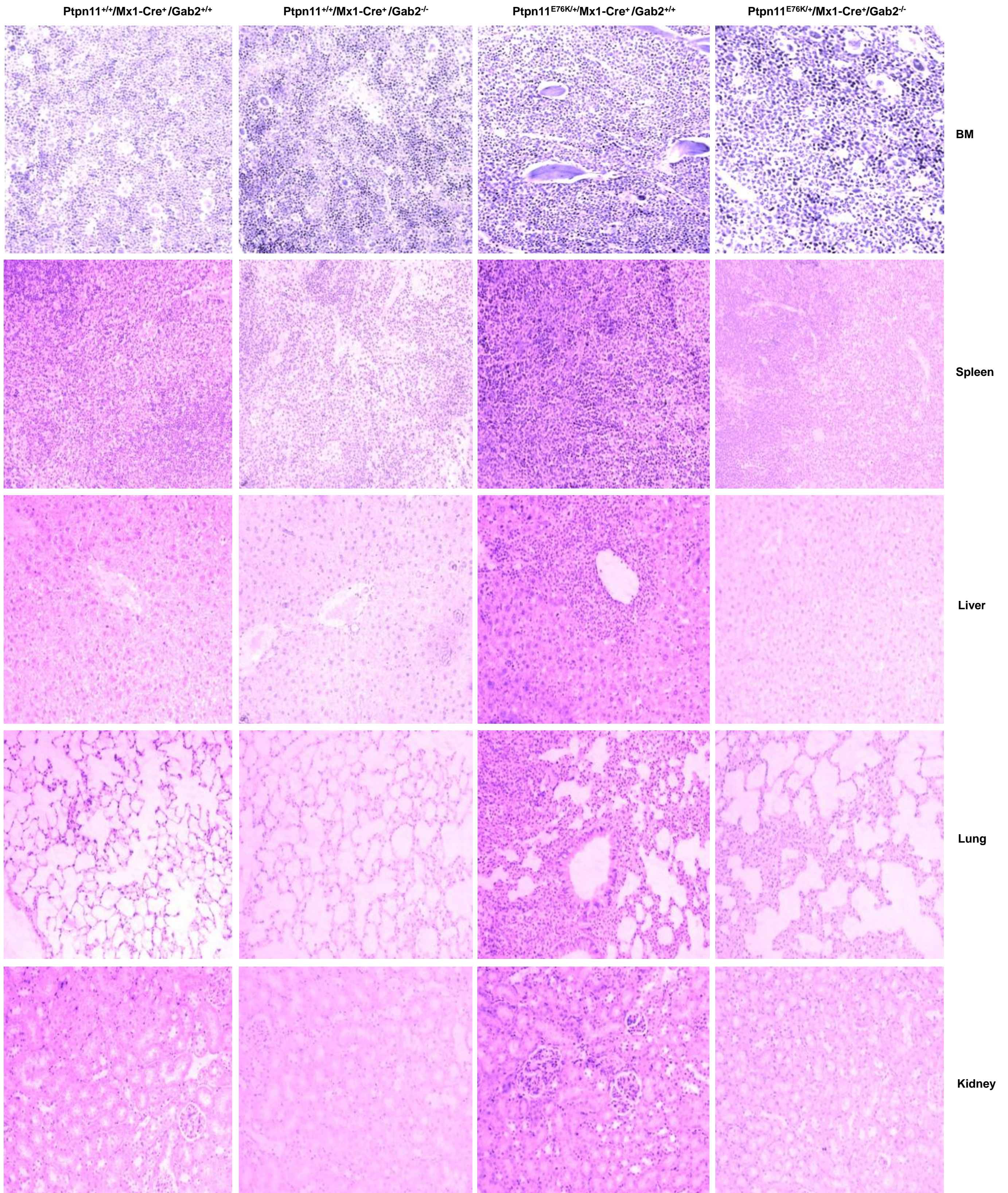
Suppl. Table 1

Peripheral blood hematology of Ptpn11^{E76K/+}/Gab2^{-/-} double mutant mice
(10~14 weeks after pl-pC treatment)

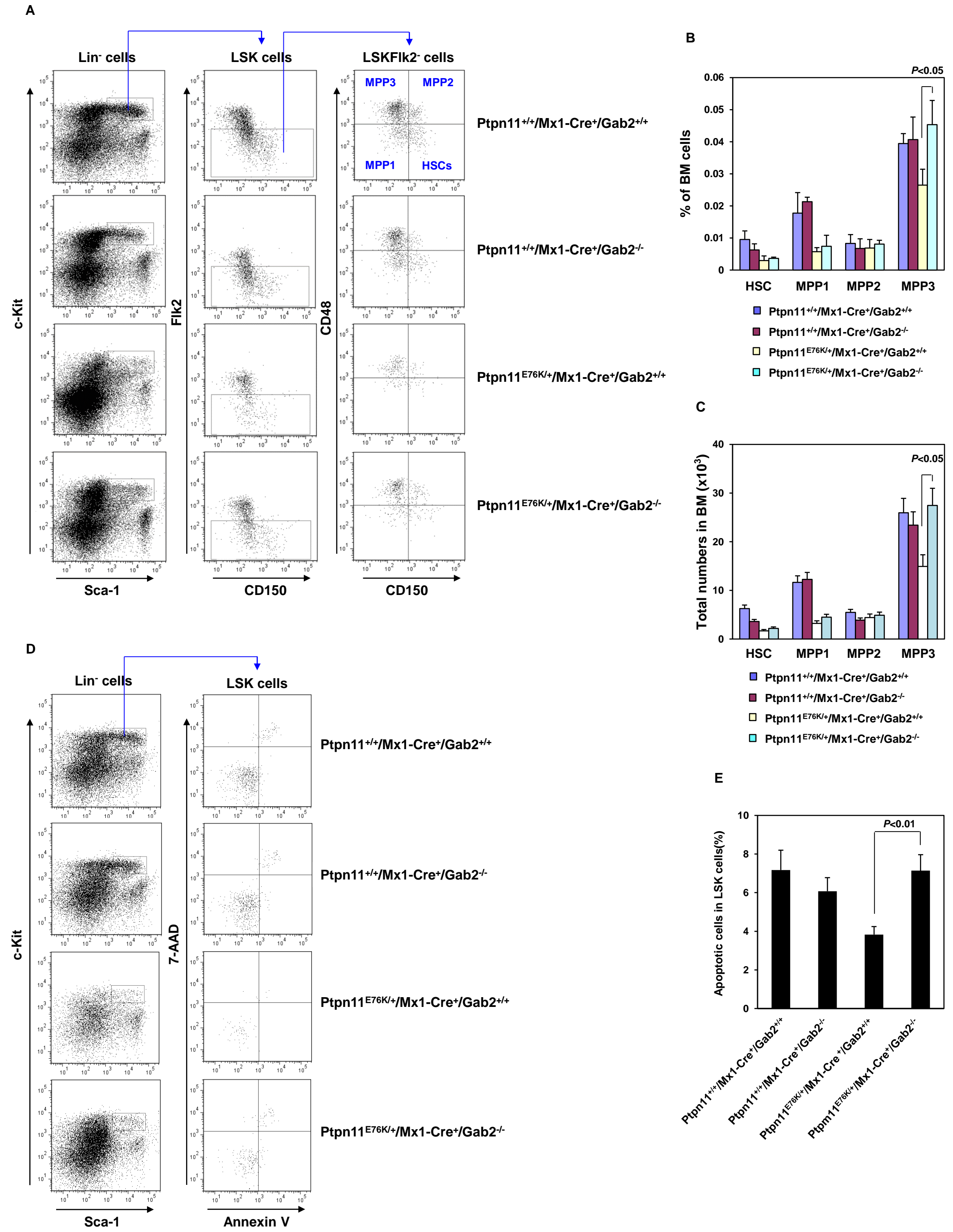
	Ptpn11 ^{+/+} /Mx1-Cre ⁺ /Gab2 ^{+/+}	Ptpn11 ^{+/+} /Mx1-Cre ⁺ /Gab2 ^{-/-}	Ptpn11 ^{E76K/+} /Mx1-Cre ⁺ /Gab2 ^{+/+}	Ptpn11 ^{E76K/+} /Mx1-Cre ⁺ /Gab2 ^{-/-}
WBC (K/ μ L)	16.35 \pm 2.71	18.88 \pm 4.42	35.55 \pm 4.51	21.01 \pm 4.83***
NE (K/ μ L)	3.63 \pm 1.04	5.90 \pm 1.61	13.17 \pm 4.49	5.42 \pm 1.31**
LY (K/ μ L)	10.92 \pm 2.85	10.91 \pm 3.04	18.50 \pm 4.91	13.94 \pm 2.48
MO (K/ μ L)	1.28 \pm 0.57	1.41 \pm 0.56	3.31 \pm 1.77	1.95 \pm 0.45
NE (%)	21.68 \pm 5.86	28.19 \pm 2.76	36.48 \pm 4.64	26.00 \pm 5.38**
LY (%)	65.97 \pm 7.49	57.94 \pm 4.50	50.28 \pm 6.22	62.27 \pm 3.93**
RBC (M/ μ L)	10.32 \pm 0.38	11.19 \pm 0.94	9.74 \pm 0.71	11.88 \pm 1.47**
Hb (g/dL)	15.33 \pm 0.83	16.04 \pm 0.86	13.72 \pm 0.68	15.69 \pm 2.70
HCT (%)	45.9 \pm 6.12	48.89 \pm 3.15	42.54 \pm 2.37	49.33 \pm 6.74*
MCV (fL)	46.05 \pm 1.90	43.89 \pm 3.83	43.46 \pm 1.71	41.47 \pm 1.42
MCH (pg)	15.03 \pm 0.81	14.74 \pm 0.65	14.29 \pm 0.71	13.34 \pm 0.91
MCHC (g/dL)	32.06 \pm 1.87	32.87 \pm 1.58	32.60 \pm 1.36	31.66 \pm 1.76
RDW (%)	17.78 \pm 0.37	18.20 \pm 0.63	19.81 \pm 0.68	19.57 \pm 0.69
PLT (K/ μ L)	649.91 \pm 167.44	1008.86 \pm 387.91	652.82 \pm 165.25	605.86 \pm 197.27

Peripheral blood was analyzed by Hemavet 950F. Data are presented as mean \pm S.D. Statistical significance between Ptpn11^{E76K/+}/Mx1-Cre⁺/Gab2^{-/-} and Ptpn11^{E76K/+}/Mx1-Cre⁺ mice was determined by unpaired, two-tailed Student's *t* tests. ***, *P*<0.0001; **, *P*<0.001; *, *P*<0.01.

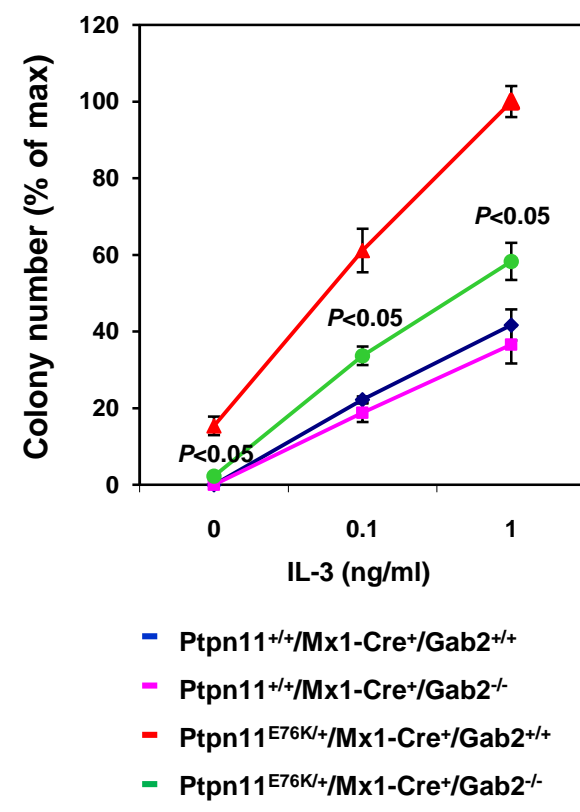
Suppl. Fig. 1



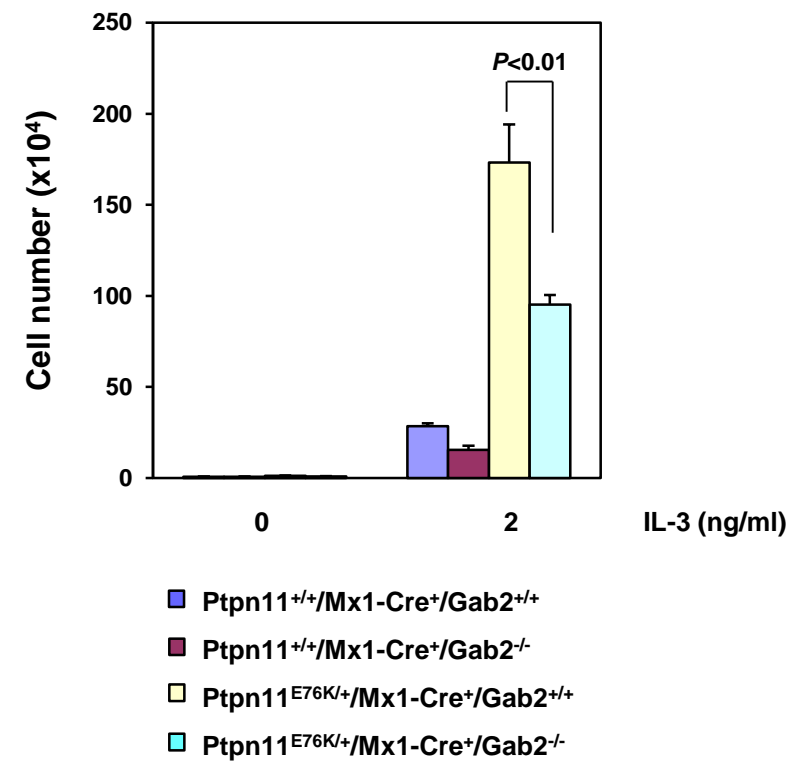
Suppl. Fig. 2



Suppl. Fig. 3



Suppl. Fig. 4



Suppl. Fig. 5

