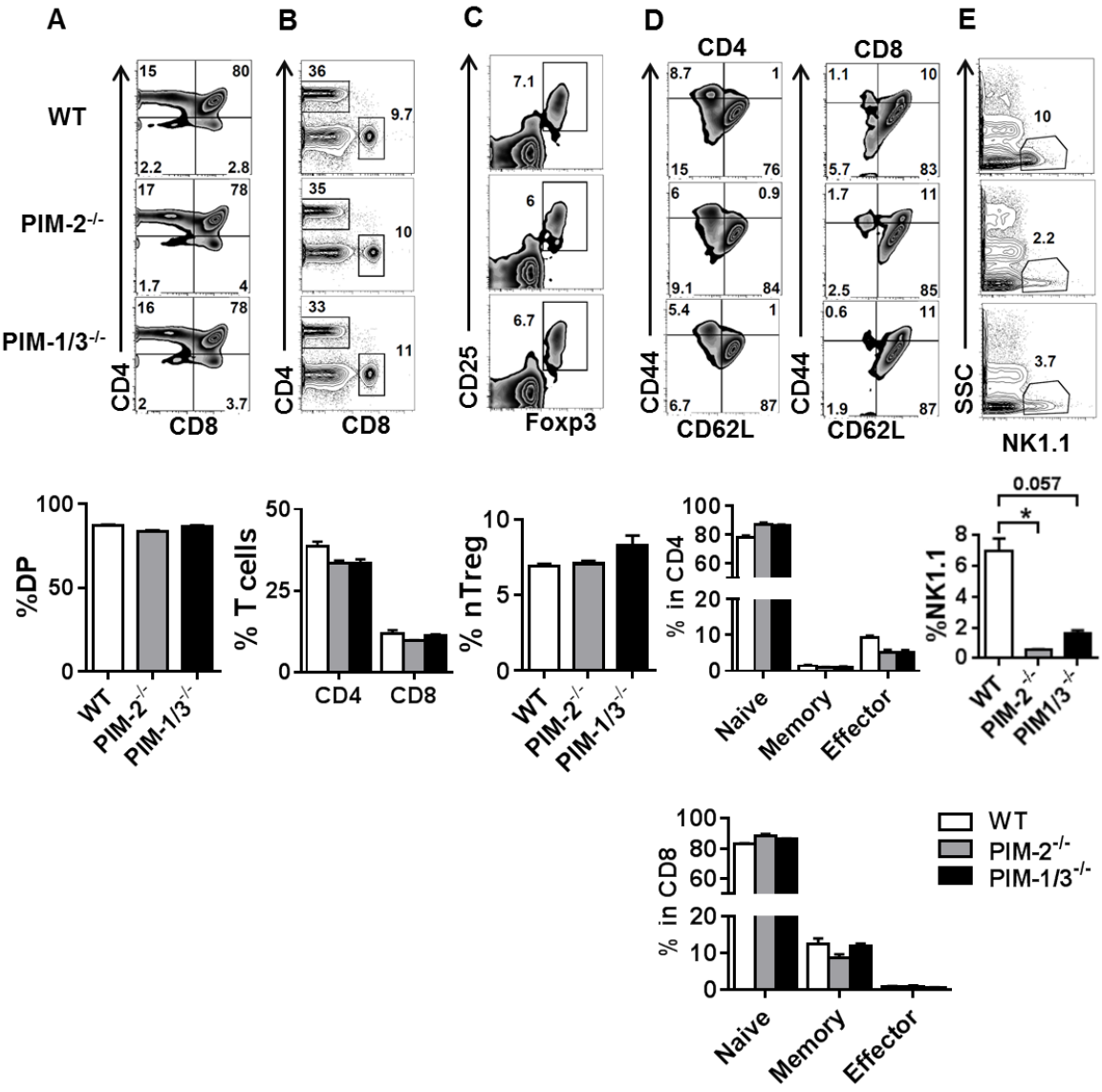
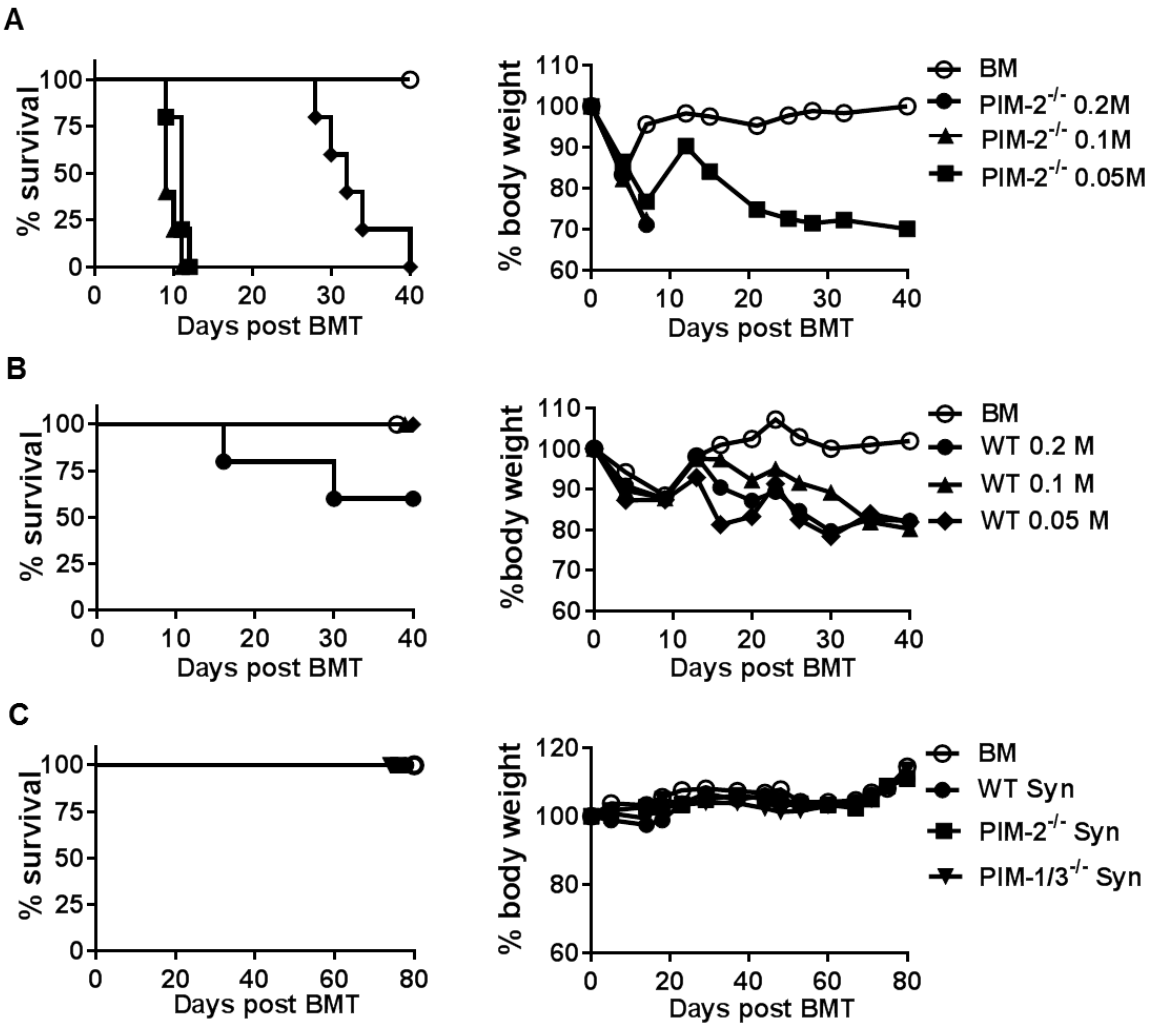
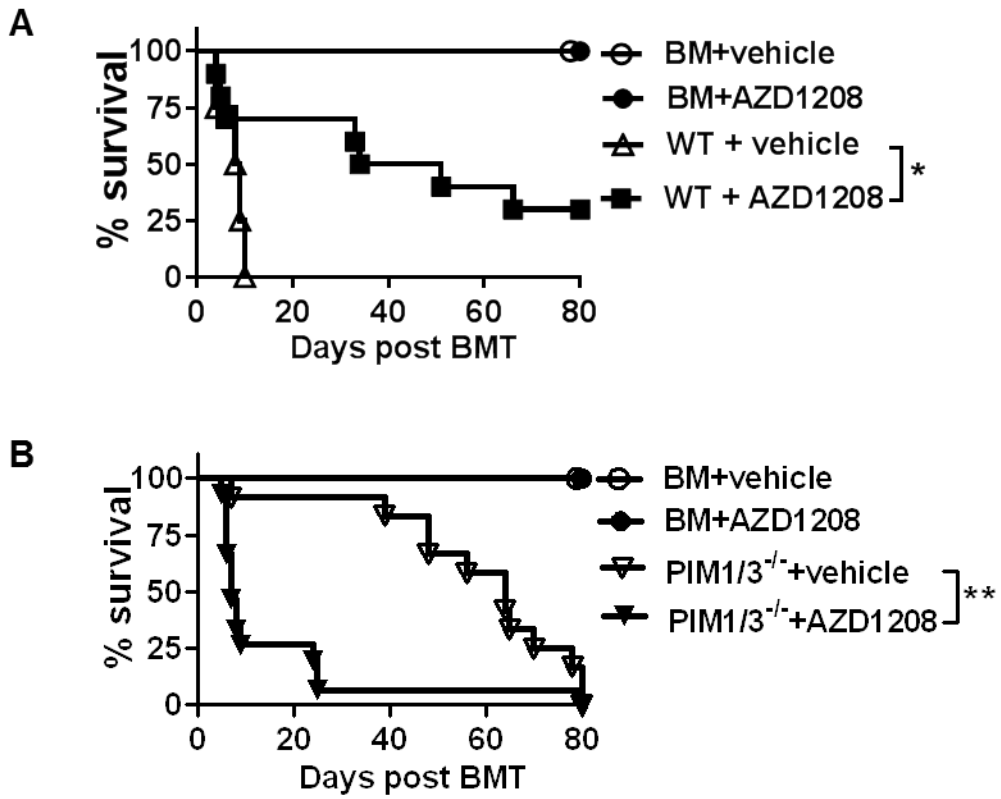
Supplemental data



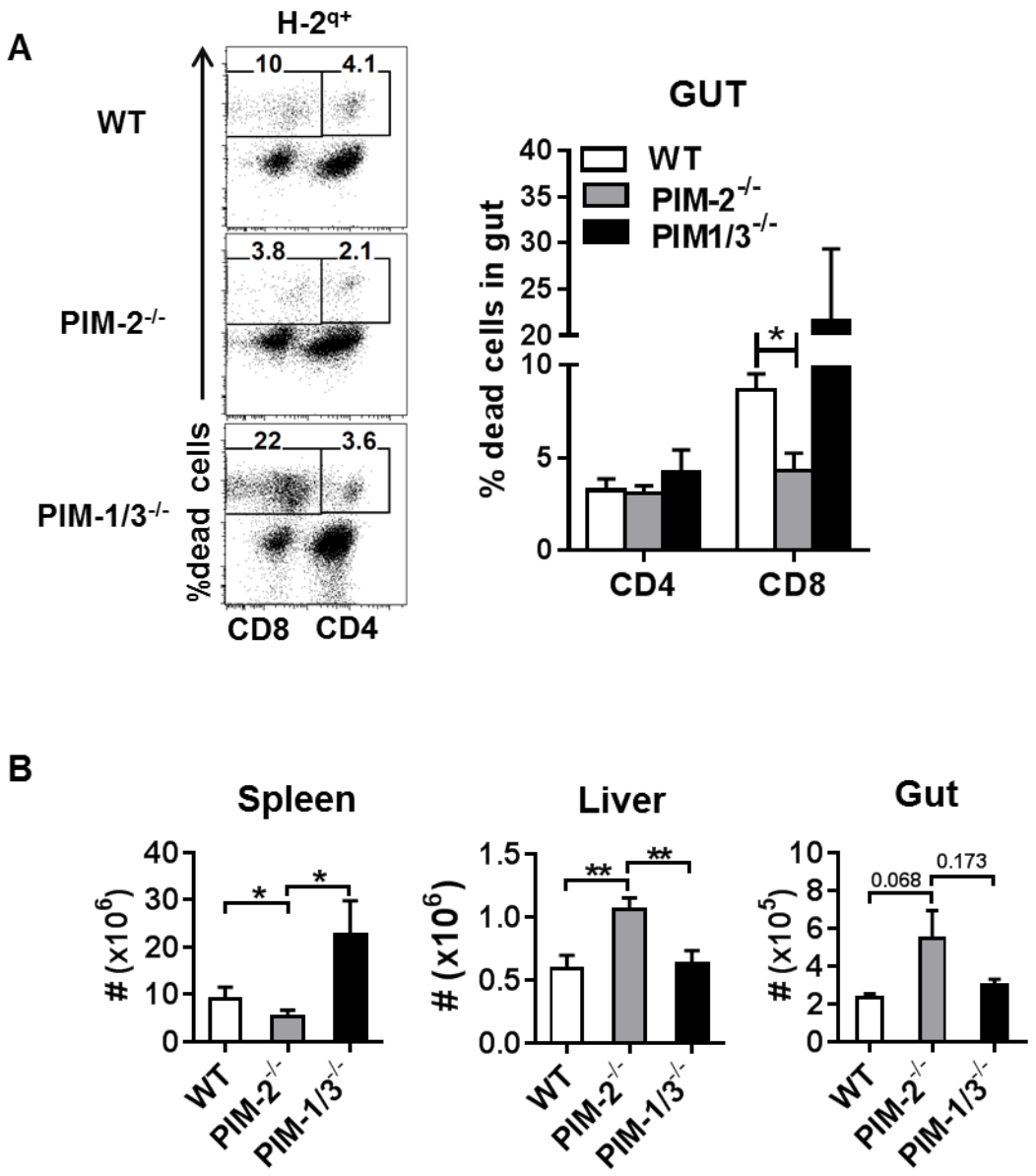
Supplemental Figure 1. T-cell phenotypes of PIM-deficient mice. (**A**) Percentages of double $CD4^+CD8^+$ in the thymus (**B**) Percentages of CD4 and CD8 T cells in the spleen of WT, PIM-$2^{-/-}$ or PIM1/$3^{-/-}$ mice. (**C**) Percentages of nTreg ($CD4^+CD25^+Foxp3^+$) in spleen. (**D**) Percentages of naive, memory and effector T cells among CD4 or CD8 T cells spleen. (**E**) The frequency of $NK1.1^+$ cells in the spleen (n=4 mice/group). Data represent mean ± SEM by two-tailed Student's *t*-test, *p< 0.05.



Supplemental Figure 2. Allogeneic and syngeneic BMT with PIM-deficient T cells. (**A**-**B**) Lethally irradiated BALB/c (700 cGy) mice were transplanted with 5 × 10^6/mouse TCD-BM plus 2x10^5, 1x10^5 and 5x10^4 isolated from WT or PIM-$2^{-/-}$ mice on a FVB background. (**C**) FVB mice were lethally irradiated (1100 cGy, FVB to FVB) and transplanted with 5 × 10^6/mouse TCD-BM 2x10^6T cells isolated from WT, PIM-$2^{-/-}$ or PIM-$1/3^{-/-}$ mice on a FVB background into syngeneic FVB recipients. Survival and body weight loss were monitored (n=4-5 mice/group).

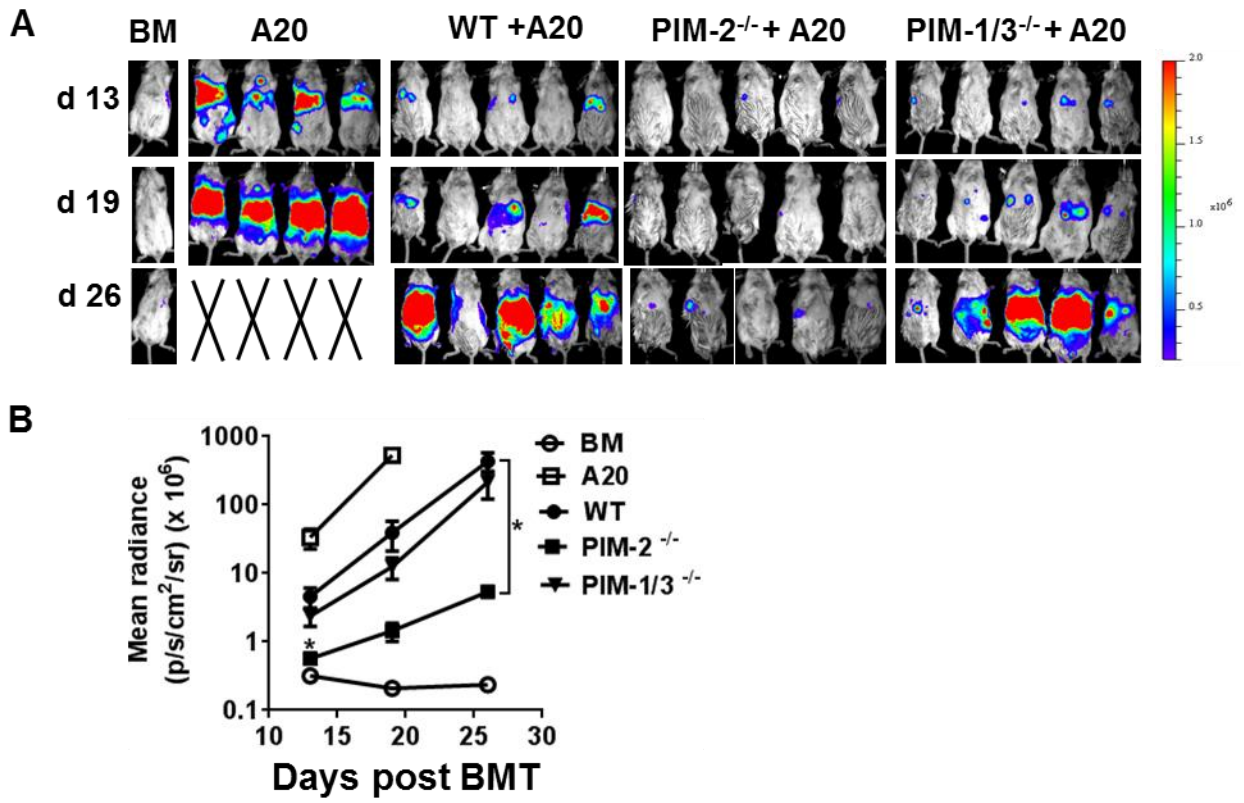


Supplemental Figure 3. The effect of pan-PIM inhibitor (AZD1208) with PIM-deficient T cells. (**A**) Lethally irradiated BALB/c (700 cGy) mice were transplanted with 5 × 10^6/mouse TCD-BM plus 5x10^5 T cells isolated from WT mice. The recipient mice were orally gavaged with either vehicle or AZD1208 15mg/kg for 14 days. The survival was monitored for 80 days (n=5-10 mice/group). (**B**) PIM-1/3$^{-/-}$ T cells (5x10^5 T cells) were transplanted together with TCD-BM into BALB/c recipients and survival of recipient mice that treated with either vehicle or AZD1208 15mg/kg for 14 days were monitored (n=10 mice/group). Significance was determined by log-rank test, *p< 0.05; **p < 0.01.

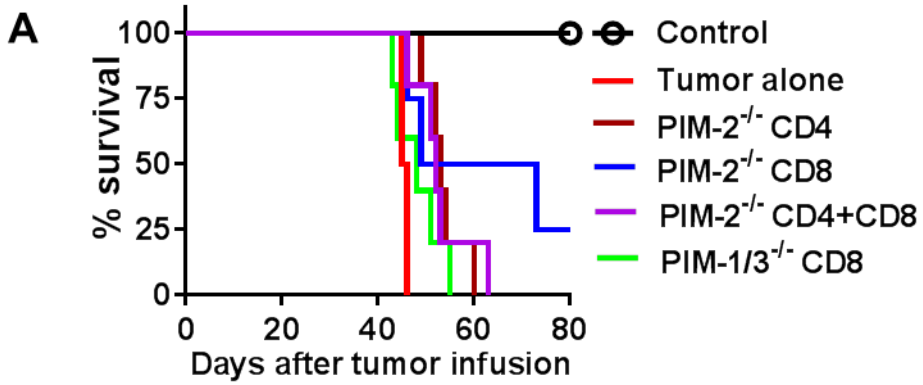


Supplemental Figure 4. PIM-2$^{-/-}$ T cells are less susceptible to apoptosis in vivo after allo-BMT. (**A**) Cells were isolated from the gut of BALB/c recipients on day 7 after allo-BMT (n=4 mice/group), and stained for expression of H-2^{q}, CD4, and CD8 with live/dead yellow dye. Percentages of dead cells were shown on gated donor CD4 and CD8 T cells. (**B**) An absolute number of donor cells recovered from spleen, liver, and gut

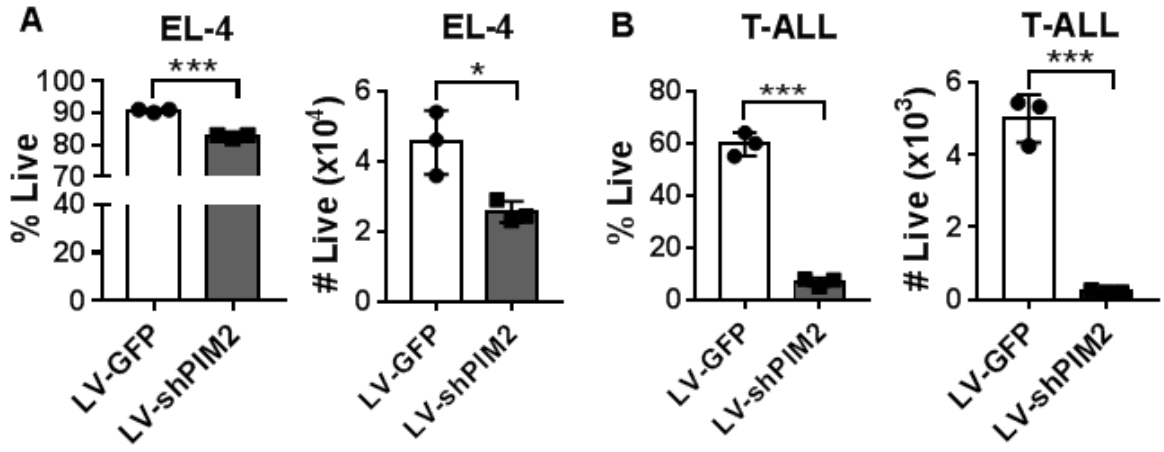
of BALB/c recipients on day 7 after allo-BMT (n=7 mice/group). Data represent mean ± SEM by two-tailed Student's *t*-test. *p< 0.05, ** p< 0.01.



Supplemental Figure 5. PIM-2$^{-/-}$ T cells mediated stronger GVL responses. (**A**) BALB/c mice were irradiated and transplanted with 5 × 10^6/mouse TCD-BM plus A20 or TCD-BM plus 50,000 WT or PIM-2$^{-/-}$ or PIM-1/3$^{-/-}$ T cells and A20 lymphoma. Tumor growth was monitored by bioluminescent imaging. (**B**) Mean radiance of tumor signals (p/s/cm^2/sr) were evaluated on day 13, 19 and 26 after allo-BMT (n=5 mice/group). Data represent mean ± SD by two-tailed Student's *t*-test. *P < 0.05.

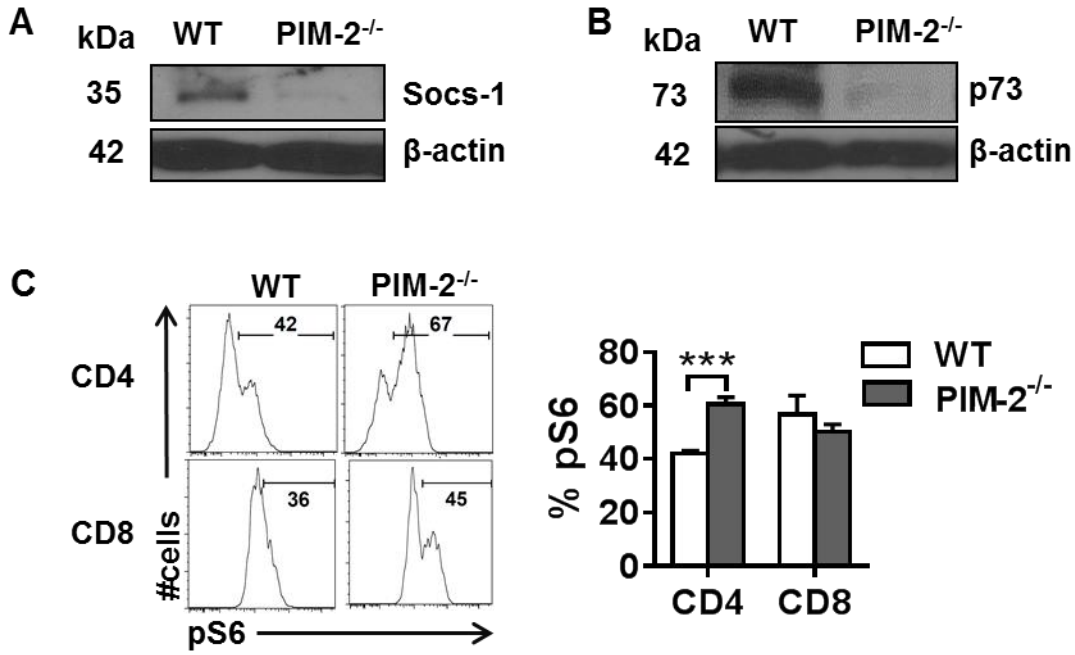


Supplemental Figure 6. The effect of PIM kinases on CD4 and/or CD8 T-cell mediated anti-tumor responses. (**A**) The TS-1 tumor was first established in WT mice for 6 days. Sub-lethally irradiation (500 cGy) was given to WT tumor-bearing mice prior to T cell transfer. $6x10^6$ of CD4 or $2x10^6$ CD8 or total T cells of PIM-$2^{-/-}$ T cells or PIM-$1/3^{-/-}$ T cells were adoptively transferred on day 7. Survival of tumor-bearing mice was shown (n=5 mice/group).



Supplemental Figure 7. PIM-2 is required for tumor cell survival. (**A**-**B**) EL-4 and T-ALL tumors were transduced with LV-GFP or LV-shPIM-2-GFP for 48 h. Cells were

harvested, stained for live/dead yellow dye and analyzed by flow cytometry for percentages of total live cells. Equal time acquisitions were used to measure an absolute number of live cells in each sample. Data represent mean ± SD by two-tailed Student's *t*-test (n=3). *P < 0.05, *** P < 0.001.



Supplemental Figure 8. Loss of PIM-2 decreases SOCS-1, p73 and increases pS6 expressions. (**A**-**B**) Western blot analysis of Socs-1 and the p73 protein expressed on donor T cells stimulated in vitro with allogeneic APC for 5 days. (**C**) Representative histogram of pS6 expression and bar graphs show the frequency of PIM-2$^{-/-}$ T cells stimulated in vitro with allogeneic APC for 5 days (n=4). Data represent mean ± SEM by two-tailed Student's *t*-test. *** P < 0.001.