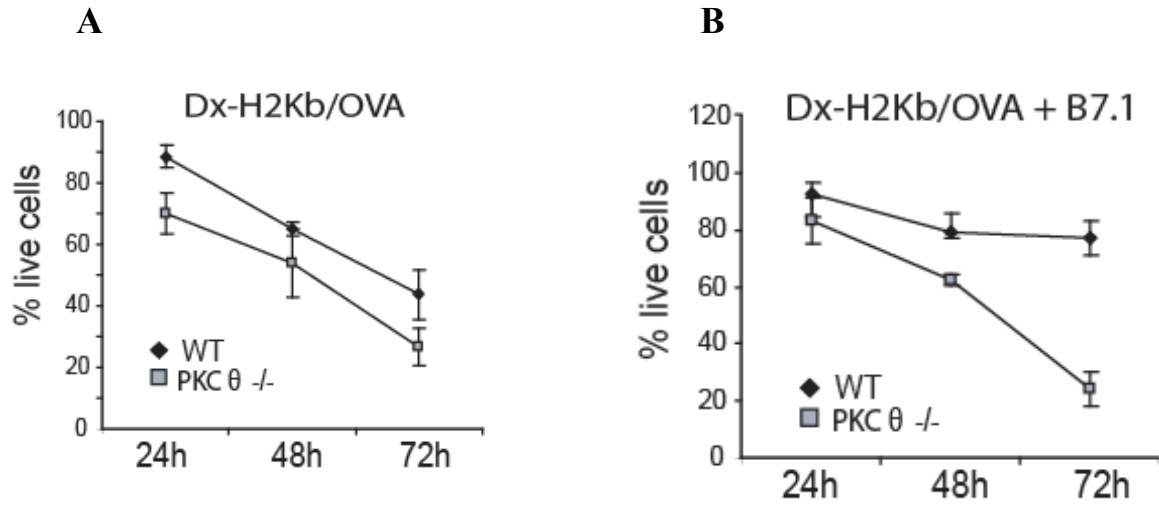
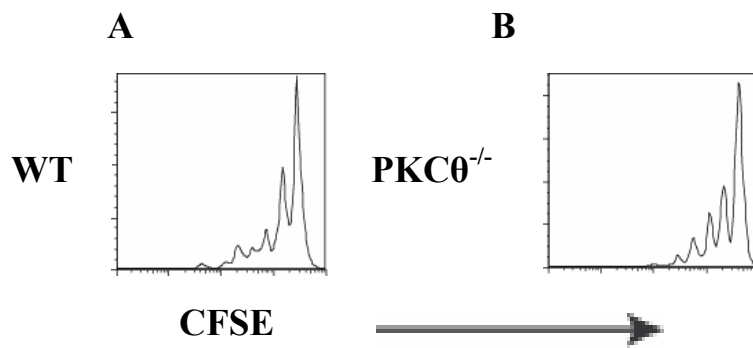


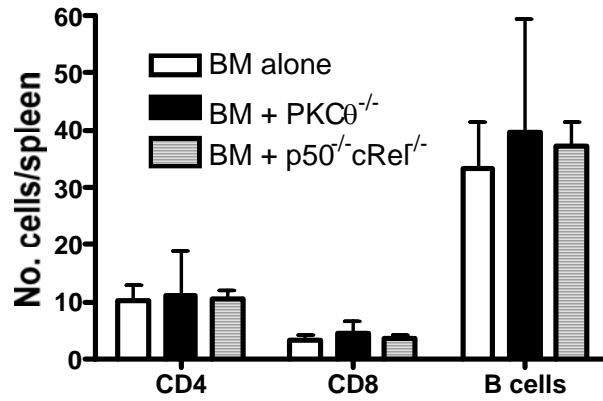
Supplemental Figures Legends



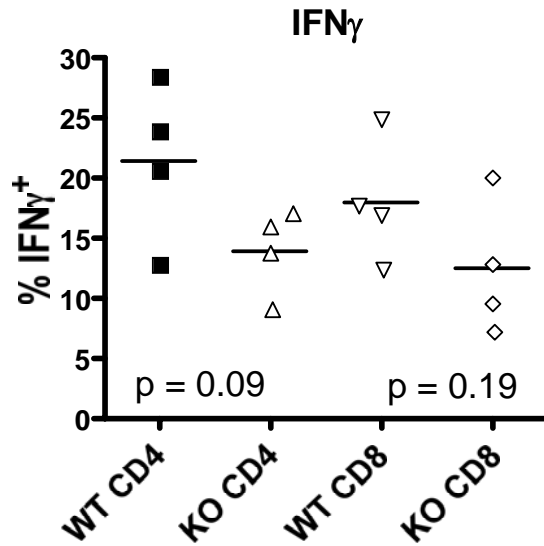
S. Fig. 1. Decreased viability of PKC $\theta^{-/-}$ CD8 T cells following activation. WT and PKC $\theta^{-/-}$ CD8 T cells were stimulated with Dx-H2Kb/OVA \pm B7.1 coated microspheres (pulsed with 0.1 μ M OVAp) and their viability was monitored using trypan blue exclusion over a period of 72h. The viability of PKC $\theta^{-/-}$ cells was lower than WT cells in the presence of OVA alone (A) or in the presence of OVA and B7.1 (B). The exclusion of trypan blue was scored from triplicate wells and reported as the percent of live cells \pm SD.



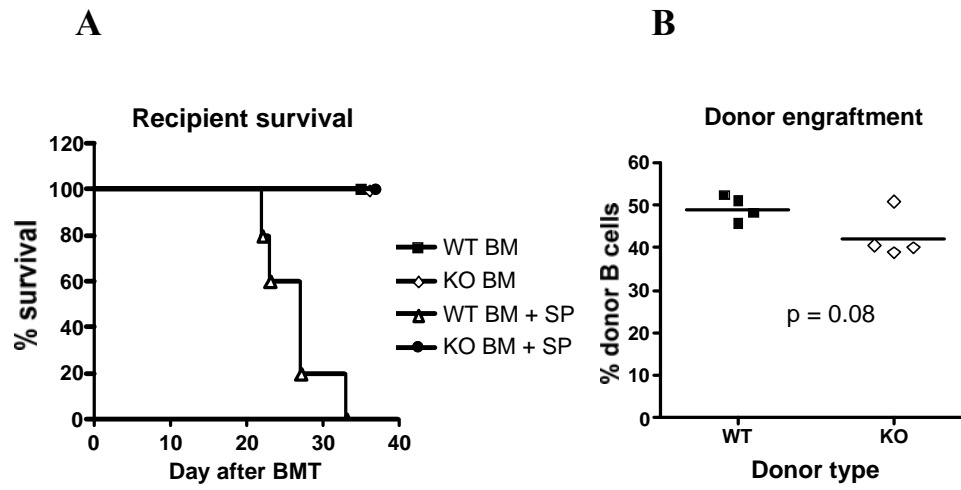
S. Fig. 2. Homeostatic proliferation of WT and PKC $\theta^{-/-}$ T cells. Homeostatic proliferation of WT (A) or PKC $\theta^{-/-}$ (B) total T cells was determined by CFSE dilution 4 days after transfer into lethally irradiated syngeneic B6 recipient mice.



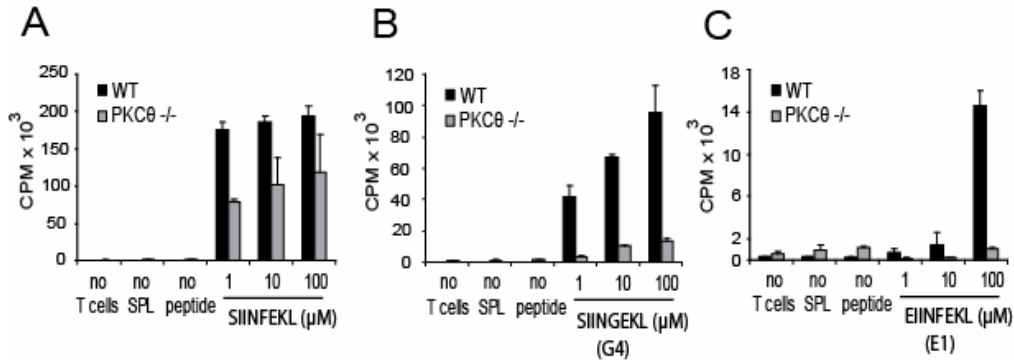
S. Fig. 3. T and B cell reconstitution post-BMT. 2×10^6 MACS-purified B6 WT, PKC $\theta^{-/-}$, or NF- κ B p50 $^{-/-}$ cRel $^{-/-}$ CD4 $^{+}$ and CD8 $^{+}$ donor T cells were injected *i.v.* into lethally irradiated (900 cGy) BALB/c recipient mice, along with T cell depleted bone marrow cells. Upon completion of the experiment on day 100, BM donor T and B cell reconstitution was measured in recipient spleens.



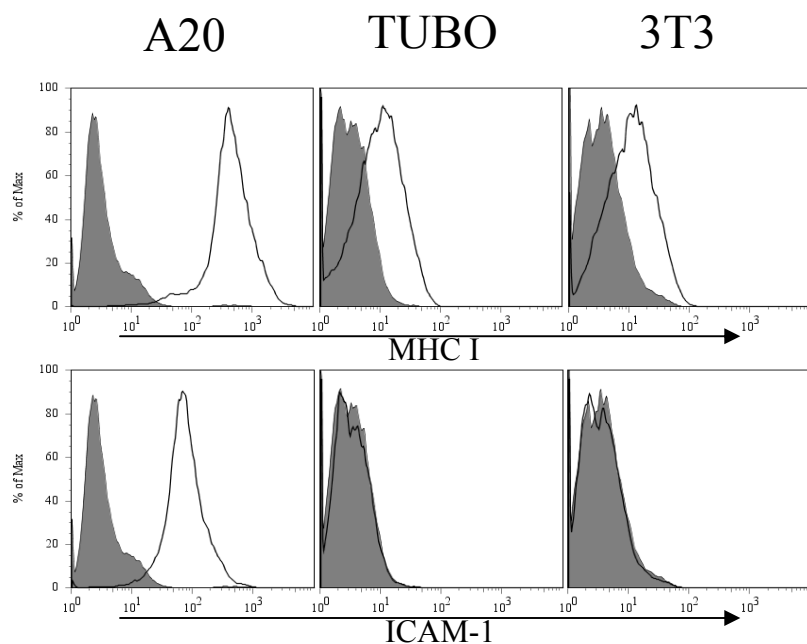
S. Fig. 4. Effect of PKC θ deficiency on T cell differentiation. 2×10^6 MACS-purified B6 WT or PKC $\theta^{-/-}$ CD4 $^{+}$ and CD8 $^{+}$ donor T cells were injected *i.v.* into lethally irradiated (850 cGy) BALB/c recipient mice. Seven days after cell transfer, spleens were harvested from each of the recipients (n = 4). Spleen cells were stained for surface expression of CD4 and H2 b and intracellular expression of IFN γ and IL-4 (not shown). Percentage of cytokine expressing cells in individual mice is shown on gated CD4 (CD4 $^{+}$ H2 $^{b+}$) or CD8 (CD4 $^{-}$ H2 $^{b+}$) T cells.



S. Fig. 5. Alloreactivity using PKC $\theta^{-/-}$ splenocytes and TCD-BM cells. Lethally irradiated BALB/c mice were injected with TCD-BM (BM) alone or BM + 20×10^6 splenocytes from WT or PKC $\theta^{-/-}$ (KO) donors. Percentage survival of recipient mice (4-6 per group) is shown (A). (B) Peripheral B cell engraftment in recipients transplanted with WT or PKC $\theta^{-/-}$ (KO) BM cells (4 mice per group).



S. Fig. 6. Proliferation of OT-1 T cells with different affinity peptide ligands. Syngeneic APCs were co-cultured with OT-1 responders in the presence of 1μM OVA₂₅₇₋₂₆₄ peptide. To measure proliferation induced by high and low-affinity peptides, 1x10⁵ WT and PKCθ^{-/-} OT-1 CD8 T cells were stimulated with 4x10⁵ irradiated splenocytes from syngeneic C57BL/6 mice, plus increasing amounts of the high-affinity OVA peptide SIINFELK (A) or its low-affinity variants SIINGEKL (G4) (B) or EIINFELK (E1) (C). Representative results from 2-3 separate experiments are shown.



S. Fig.7. Cell surface expression of MHC I (H2D^d) and ICAM-1 on A20, TUBO epithelial cell line and 3T3 fibroblasts, as indicated. Cells were labeled with FITC-MHC I (top) or FITC-ICAM (bottom.) Filled histograms depict unstained control cells; open histograms show surface expression of indicated markers.