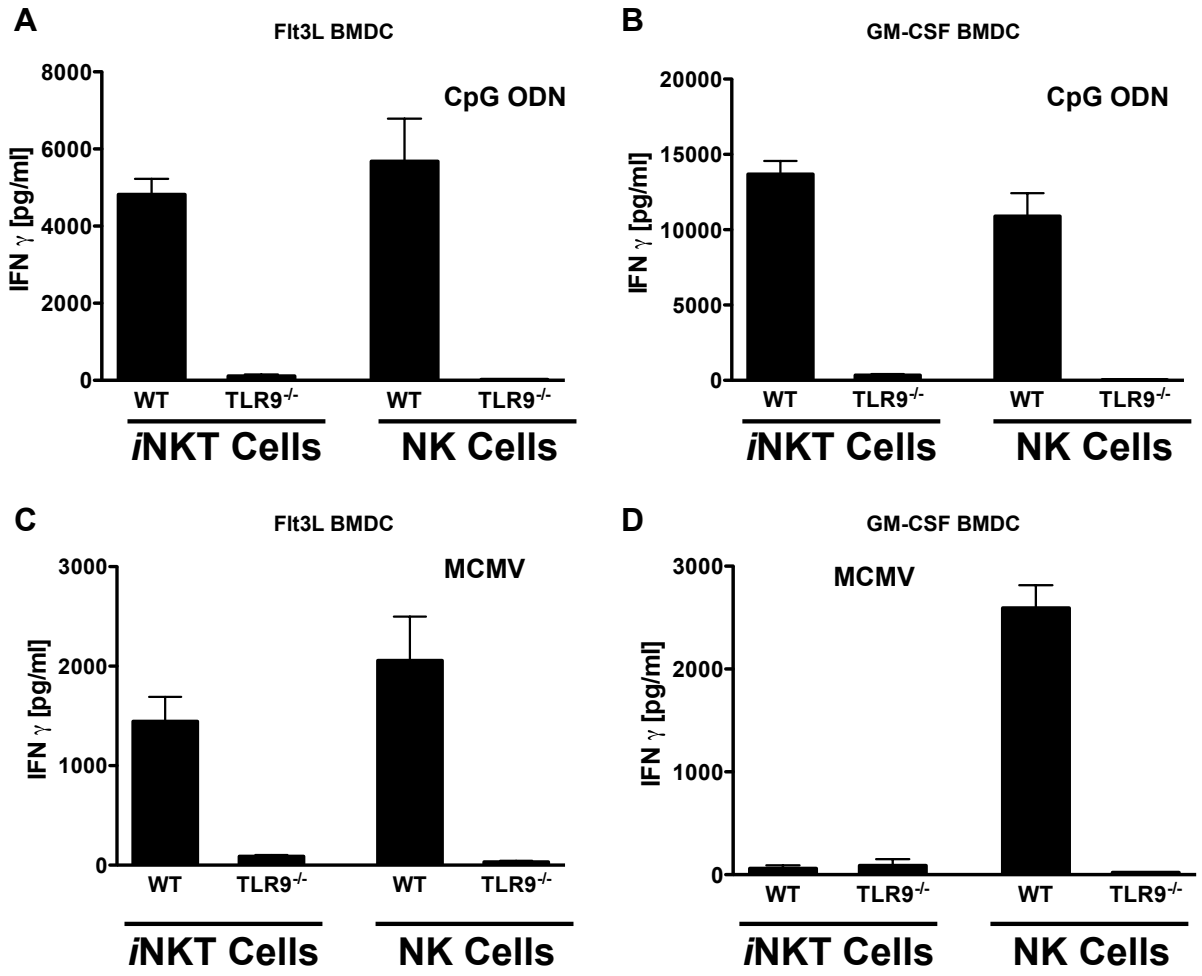
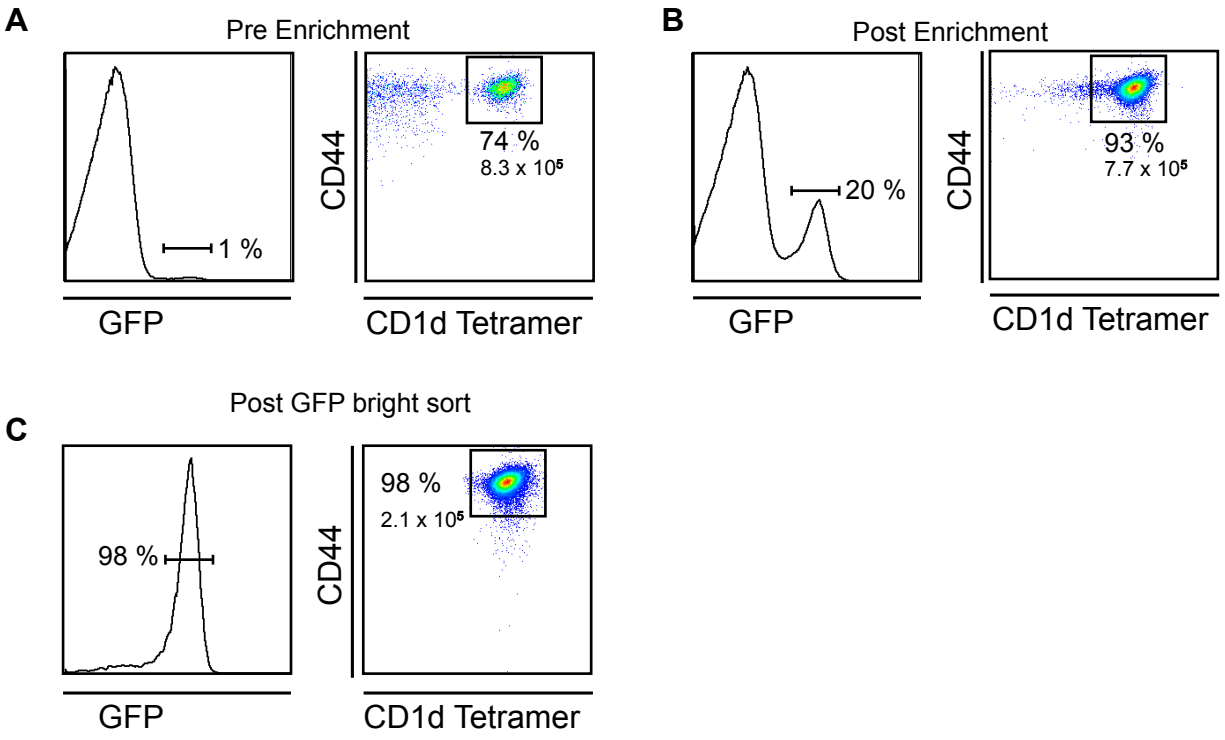


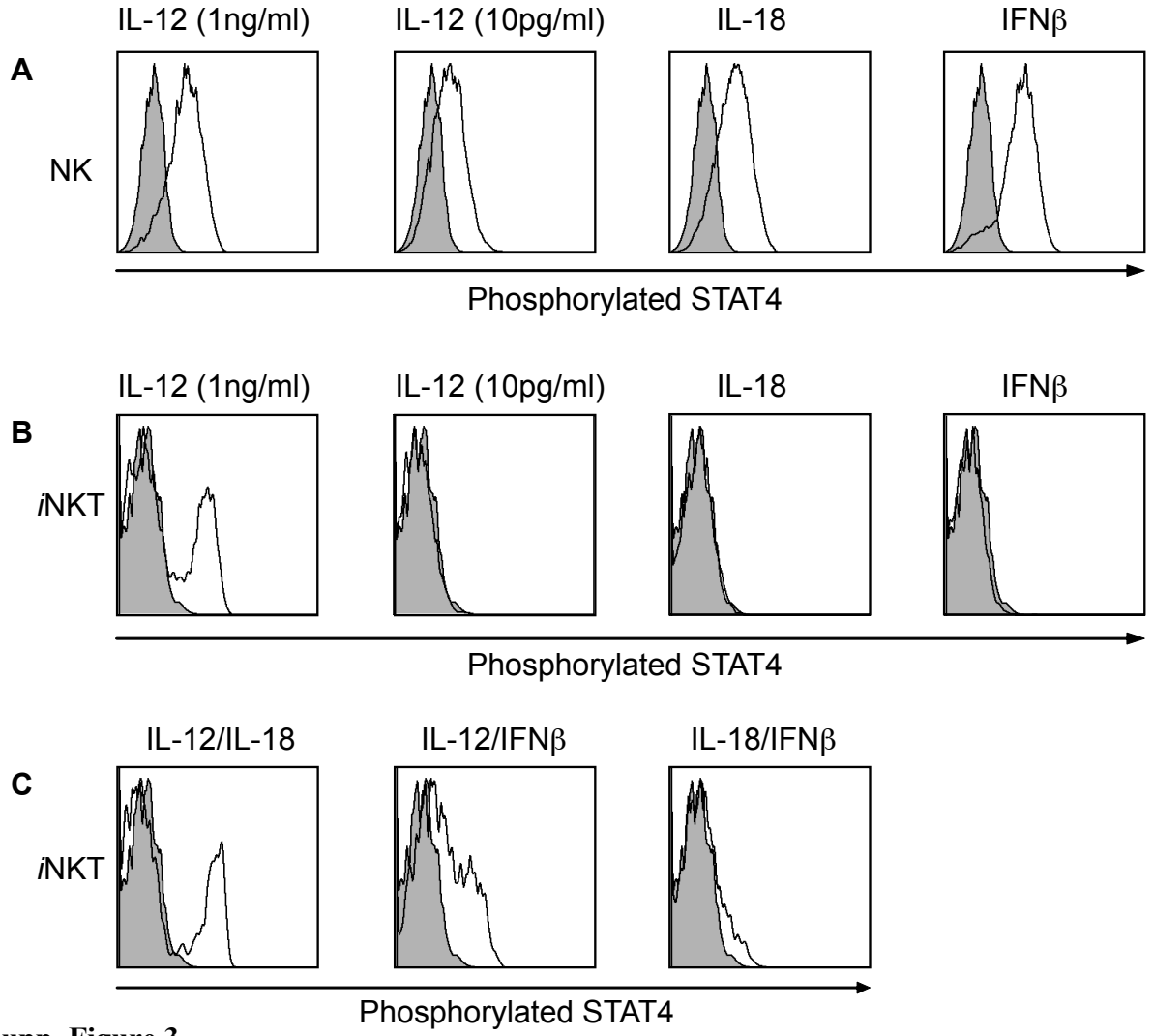
Supp. Figure 1.

IFNγ production by *i*NKT and NK cells in DC co-cultures is TLR9 dependent. Flt3L (*A*) or GM-CSF (*B*) derived BMDC from *Tlr9*$^{cpg/cpg}$ mice were treated with CpG-ODN or control treated and cultured with purified *i*NKT cells or NK cells for 48h. Flt3L (*C*) or GM-CSF (*D*) derived BMDC from *Tlr9*$^{cpg/cpg}$ were mock or MCMV infected and cultured with purified *i*NKT cells or NK cells for 48h. Supernatants were analyzed for IFNγ by ELISA, and shown is a representative experiment of 3 performed. ELISA results represent the mean of one experiment with three replicate cultures measured in triplicate. Error bars represent SEM (n = 9 for each set of BMDC).



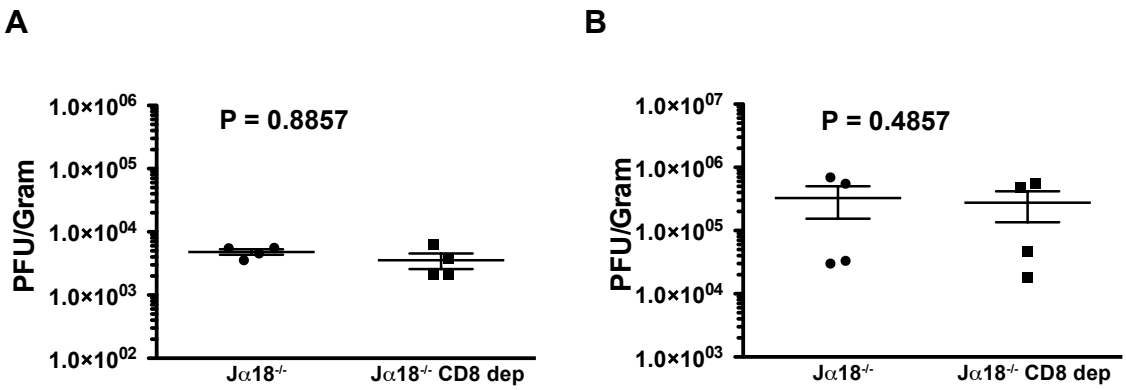
Supp. Figure 2.

No-touch isolation of *i*NKT cells. *A*, Pre-enrichment splenocytes of C57BL/6 4get mice were analyzed for eGFP$^+$ cells (*left panel*). Analysis of pre-enrichment eGFP$^+$ cells analyzed for CD44$^+$ and CD1d αGalCer tetramer$^+$ *i*NKT cells (*right panel*). *B*, Post-enrichment splenocytes of C57BL/6 4get mice analyzed for eGFP$^+$ cells (*left panel*). Analysis of post-enrichment eGFP$^+$ cells analyzed for CD44$^+$ and CD1d αGalCer tetramer$^+$ *i*NKT cells (*right panel*). *C*, Post sort analysis of GFP^{bright} splenoyctes for CD44$^+$ and CD1d αGalCer tetramer$^+$ *i*NKT cells. *A*-*C*, Percentages represent gated eGFP$^+$ and gated *i*NKT cells. Numbers represent absolute number of *i*NKT cells per spleen. Data are representative of 8 independent experiments.



Supp. Figure 3.

NK and *i*NKT cell STAT4 activation. Splenocytes and liver mononuclear cells (data not shown) were isolated from naïve C57BL/6 4get mice and left untreated or stimulated with IFNβ, IL-12, or IL-18 alone (*A and B*) or in combination (*C*) for 1 hour and cells were collected and stained to detect intracellular STAT4 phosphorylation. (*A-B*), Flow cytometric analysis of (*A*) $CD19^-$, $CD3^-$, $NK1.1^+$ NK cells (*B*) or $CD19^-$, $CD8^-$, $CD3^+$ $CD44^+$ GFP^+ *i*NKT cells left untreated (shaded histogram) or cultured with hi dose IL-12 (1ng/ml), IL-18 (1ng/ml) or IFNβ (1000U/ml) indicated by the solid line. (*C*), Flow cytometric analysis of $CD19^-$, $CD8^-$, $CD3^+$ $CD44^+$ GFP^+ *i*NKT cells left untreated (shaded histogram) or cultured with low dose of IL-12 (10pg/ml), IL-18 (10pg/ml), and IFNβ (10U/ml) as indicated and represented by the solid line. Data are representative histogram plots of 3 mice per condition cultured in triplicated.



Supp. Figure 4.

Depletion of $CD8^+$ T cells does not alter day 4 MCMV innate defenses. Groups of BALB/c $J\alpha18^{-/-}$ *i*NKT cell-deficient mice were depleted of $CD8^+$ T cells or not, and subsequently infected with MCMV. MCMV replication levels in the liver *(A)*, and spleen *(B)*, are shown day 4 post infection. Results are representative of 4 mice per group. Error bars represent SEM (n=4 for each group).