



**SUPPLEMENTAL FIGURE 1.** *Nlr5* plays critical roles in MHC class I expression and host defense against *L. monocytogenes* infection but not TLR responses. **(A)** Peritoneal cavity cells, bone marrow-derived dendritic cells (BMDC) and macrophages (BMDM) from *Nlr5* *+/+* (grey line) and *Nlr5* *-/-* (black line) mice were analyzed by flow cytometry for the expression of MHC class I (H2-K/D/L) gated on B220<sup>+</sup>, F4/80<sup>+</sup> or CD11c<sup>+</sup> cells (left panel). Shaded regions represent the isotype control. Bar graphs (right panel) represent the mean and SEM of the corresponding MFI (n = 3 for each indicated genotype). \*p < 0.05; ns, not significant. **(B)** Expression of MHC class I (*H2-K<sup>b</sup>*) in BMDC and BMDM of *Nlr5* *+/+*, *Nlr5* *+/+* and *Nlr5* *-/-* mice were determined by qPCR analysis using gene-specific primers. n = 3 for each indicated genotype. Error bars represent  $\pm$  SEM. \*p < 0.05. **(C)** *Nlr5* *+/+* and *Nlr5* *-/-* mice were infected with *L. monocytogenes* (1 X 10<sup>4</sup> CFU). Untreated splenocytes and hepatic leukocytes isolated from *Nlr5* *+/+* and *Nlr5* *-/-* mice at day 6 after infection were analyzed by flow cytometry for the expression of IFN- $\gamma$  gated on CD8<sup>+</sup> cells. Data are representative of two independent experiments. **(D)** BMDM from *Nlr5* *+/+* (n=3) and *Nlr5* *-/-* mice (n=3) were stimulated with LPS (10 ng/ml) for 16 hours. Transcript levels of IL-6, TNF- $\alpha$ , IL-12p40 and IL-1 $\beta$  were analyzed by qPCR using the indicated gene-specific primers. Error bars represent  $\pm$  SEM. ns, not significant. **(E)** BMDM from *Nlr5* *+/+* (n=3) and *Nlr5* *-/-* mice (n=3) were stimulated with LPS, CpG DNA (10  $\mu$ M) or poly(I:C) (50  $\mu$ g/ml) for 16 hours. Cell free supernatants were analyzed for the level of IL-6 and TNF- $\alpha$  by ELISA. Error bars represent  $\pm$  SEM. ns: not significant.