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## Supplemental Data

# Toll-Like Receptor 6 Drives Differentiation of Tolerogenic Dendritic Cells and Contributes to LcrV-Mediated Plague Pathogenesis

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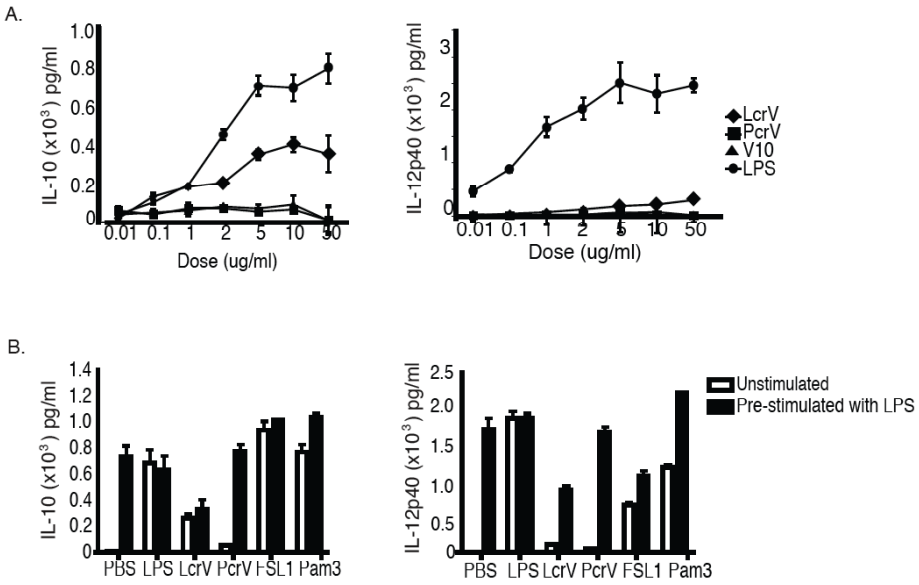


**Figure S1.** (A) Dose response of IL-10 (left) and IL-12p40 (right) treated with increasing concentration of LcrV, PcrV, V10 and LPS. (B) BMDC unstimulated or pre-stimulated with 0.1ug/ml LPS and after 2 hours pulsed with LPS, LcrV, PcrV, FSL1 or Pam3CysK4 (Pam3). IL-10 and IL-12p40 were measured 18 hours later by ELISA.

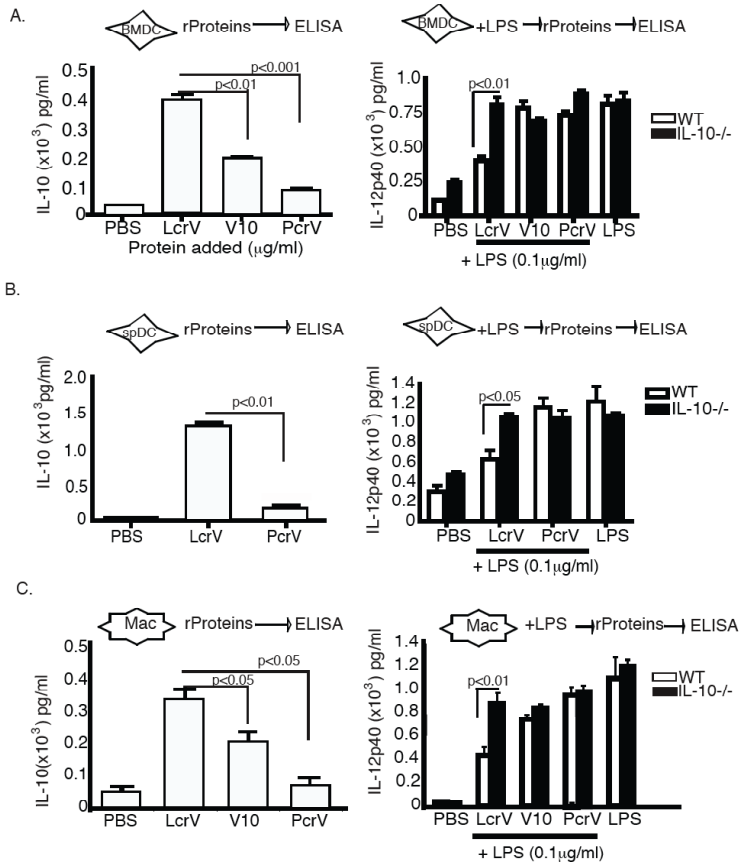


**Figure S2.** (A) Left, dendritic cells were isolated from spleen of C57BL/6 mice and treated directly with 10ug/ml LcrV or PcrV and supernatants were tested for IL-10 or first pulsed with 0.1ug/ml LPS for 2 hours and then pulsed with LcrV or PcrV and measured for IL-12p40 (right). (B) Same as described above, but cells were bone marrow derived DC and V10, an LcrV deletion mutant, was also screened. (C) same as described in (A) but peritoneal macrophages were used as the responding cell type. *, $p<0.05$ One-way ANOVA

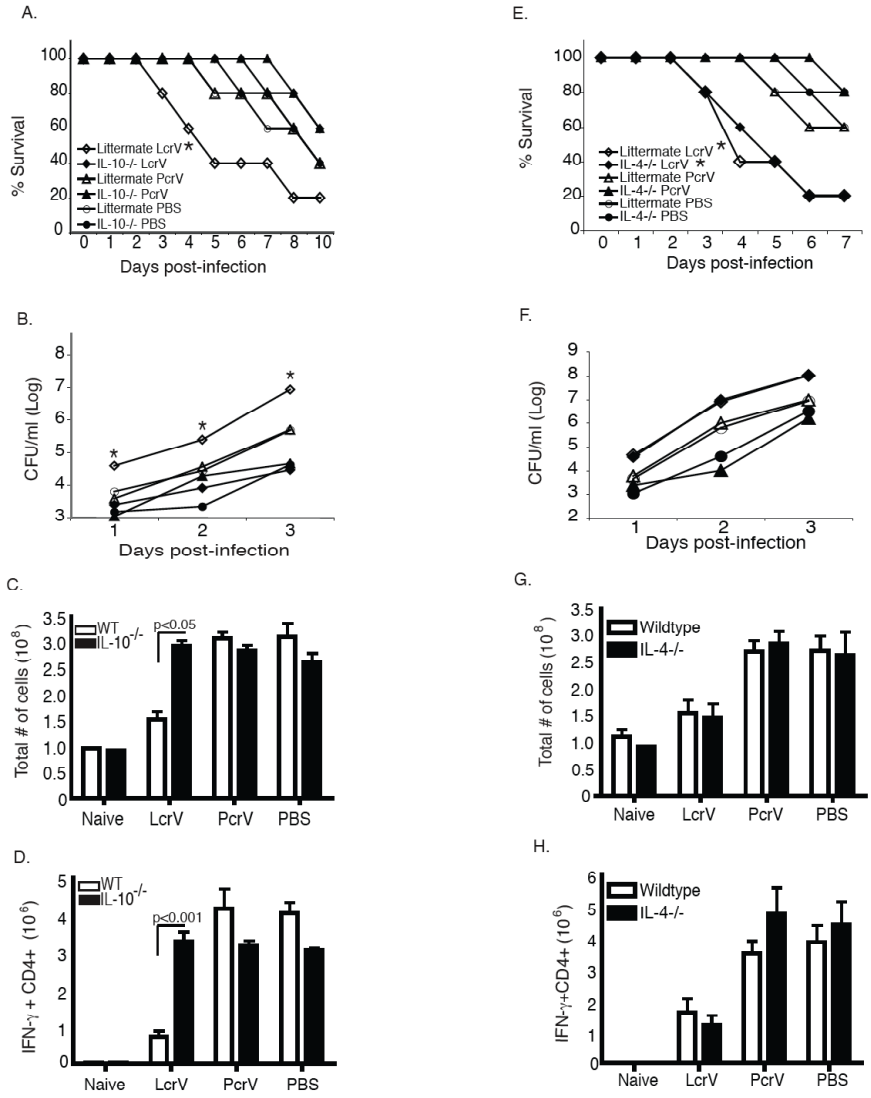


**Figure S3.** IL-10-/-, IL-4-/- and littermate controls were infected treated with 50ug/ml LcrV or PcrV and infected intravenously with $10^3$cfu Listeria monocytogenes. (A) Survival curve of IL-10-/- mice, p=0.002 by Wilcoxon log rank test. (B) Bacterial load in spleen was determined at days 1-3 after infection. (C) At day 6 splenic cell count and (D) intracellular IFN-γ levels were measured. (E) IL-4-/- survival curve. (F) Bacterial load in IL-4-/- mice after infection with Listeria and treatment with LcrV. (G) and (H) are cell counts and IFN-γ, respectively. Two-way ANOVA was used for analysis of panels

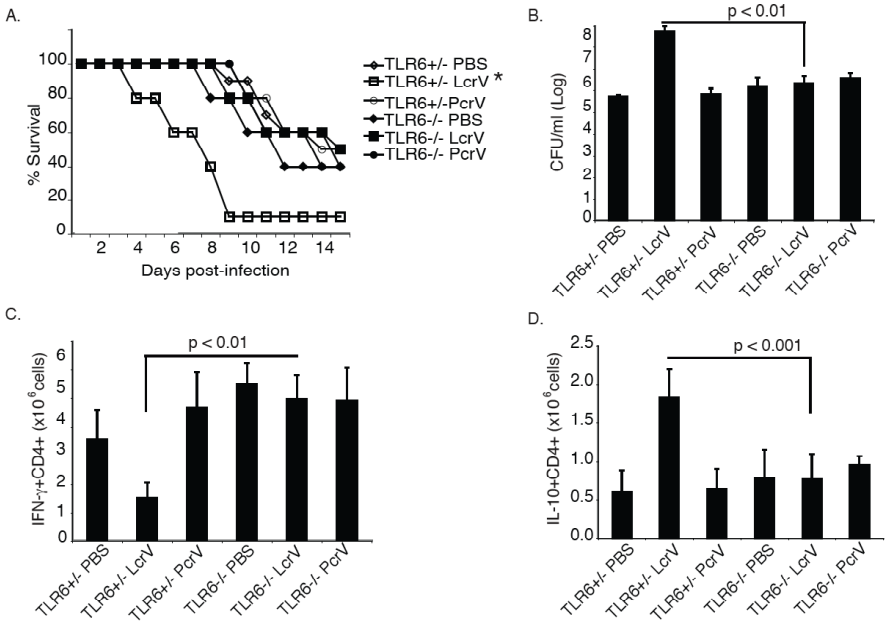


**Figure S4**. TLR6-/- and littermate controls were treated and infected as described in Supplementary Figure 4. (A) Survival curve of mice, $p<0.001$ by Wilcoxon log rank test. (B) Splenic cell count at day 6 and (C) Intracellular CD4+IFN-γ+ cells measured by flow cytometry on day 6.

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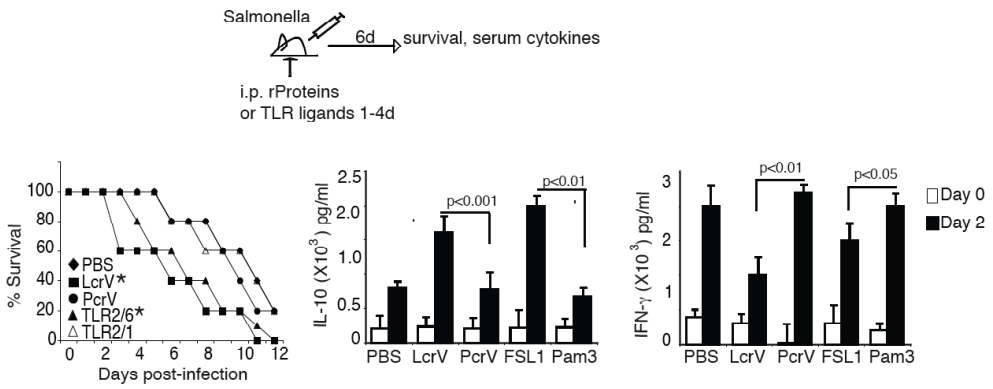


**Figure S5.** C57BL/6 mice were infected i.v. with 1000cfu *Salmonella typhimurium*. The mice were then injected with 50ug LcrV or PcrV or 10ug FSL1 or Pam3CysK4 once a day for 4 days. Survival (n=10, *left*) and serum levels of IL-10 and IFN-γ (*center* and *right*) were measured (n=5mice/group). Wilcoxon-rank test gave $p<0.005$ for survival. One-Way ANOVA was used for analysis of IL-10 (LcrV to PcrV $p=0.001$; FSL1 and Pam3CysK5 $p= 0.001$) and IFN-γ (LcrV to PcrV $p=0.01$; FSL1 and Pam3CysK5 $p>0.05$).