## Supplemental Figure 1



**Figure S1:** *C. difficile* results in rapid up-regulation of inflammatory and anti-microbial **genes.** Total cecum and colon tissues were harvested and analyzed for gene expression by qPCR following *C. difficile* (5x10<sup>5</sup> CFU; N=4-5 per time point). Normalized to day 0 sample with GAPDH as endogenous control.



**Figure S2:** *C. difficile* leads to up-regulation of IL-17A-producing cells. Single-cell suspensions of total cecum tissue from naïve (day 0) and day 4-infected mice (5x105 CFU) were stimulated with PMA/ionomycin *in vitro* followed by intracellular staining and analyzed by flow cytometry. Plots shown are gated on live CD45+ cells (data representative of two experiments).



**Figure S3:** Neutrophil recruitment is not attenuated during *C. difficile* of IL-17Adeficient mice. (A) Neutrophil infiltration into intestine of WT and *II17a-/-* following *C. difficile* (4x10<sup>5</sup> CFU). Gated on live CD45+ CD11b+ Ly6G+ cells (data combined from two experiments; N=11 per genotype). (B) Percent survival of WT mice treated with isotype control (2A3) and anti-Ly6G antibody (1A8) following *C. difficile* (4x105 CFU; not significant, log-rank test).



Figure S4: Alpha-beta T cells and gamma-delta T cells in uninfected mice do not express CD69 at baseline. Surface expression of CD69 in alpha-beta T cells and gamma-delta T cells in antibiotics-treated uninfected mice. Filled histograms represent isotype control staining. Gated on live CD45+ CD3 $\epsilon$ + CD4+ TCR $\beta$ + cells or live CD45+ CD3 $\epsilon$ + TCRgamma-delta+ cells (results representative of two experiments).



Figure S5: *C. difficile*-responsive IL-17A+ gamma-delta T cells do not express typical V<sub>Y</sub> chains. Single-cell suspensions from tissues of day 4-infected mice (4x10<sup>5</sup> CFU) were stimulated with PMA/ionomycin *in vitro* followed by intracellular staining and analyzed by flow cytometry. Gated on live CD45+ CD3 $\epsilon$ + TCRgamma-delta+ cells. Y-axis labeled according to Tonegawa system (V<sub>Y</sub>1 = *Trgv*1, V<sub>Y</sub>4 = *Trgv*4, V<sub>Y</sub>5 = *Trgv*5, and V<sub>Y</sub>7 = *Trgv*7).



**Figure S6: Gating strategy for the isolation of mLN IL-17A- and IL-17A+ gammadeltaT-cells.** Mesenteric lymph nodes were harvested from day 4-infected mice (4x10<sup>5</sup> CFU), stimulated with PMA/ionomycin *in vitro* and stained by surface cytokine capture.