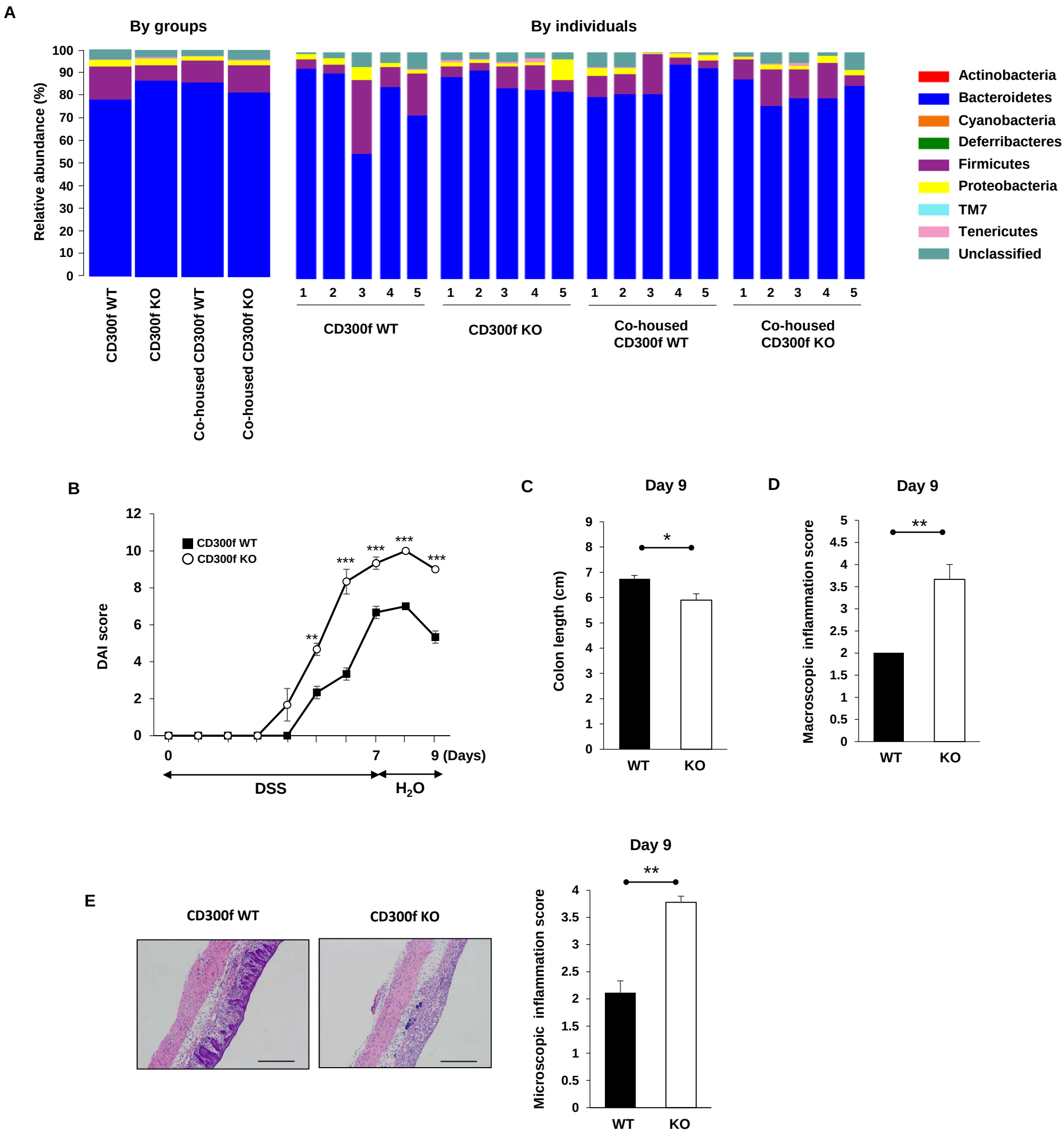
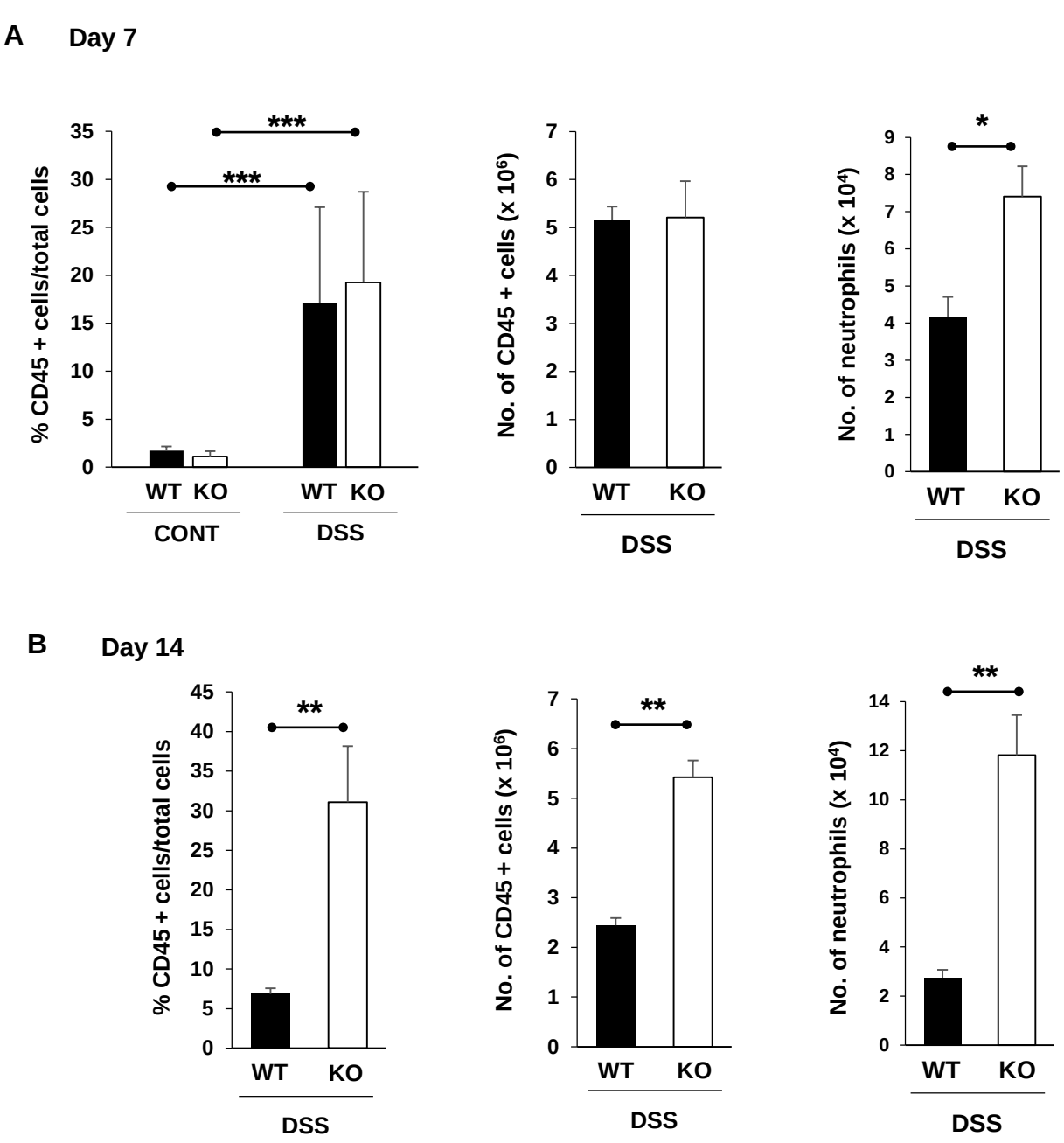
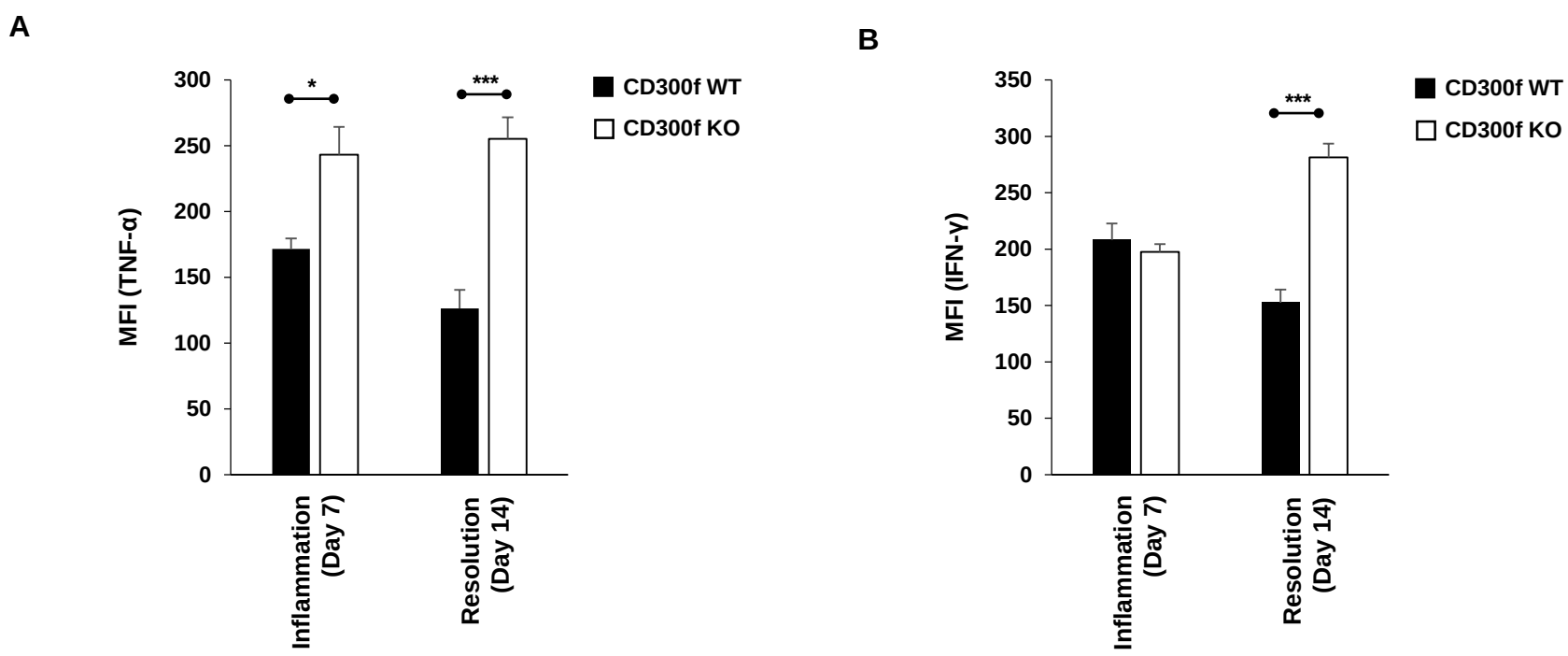


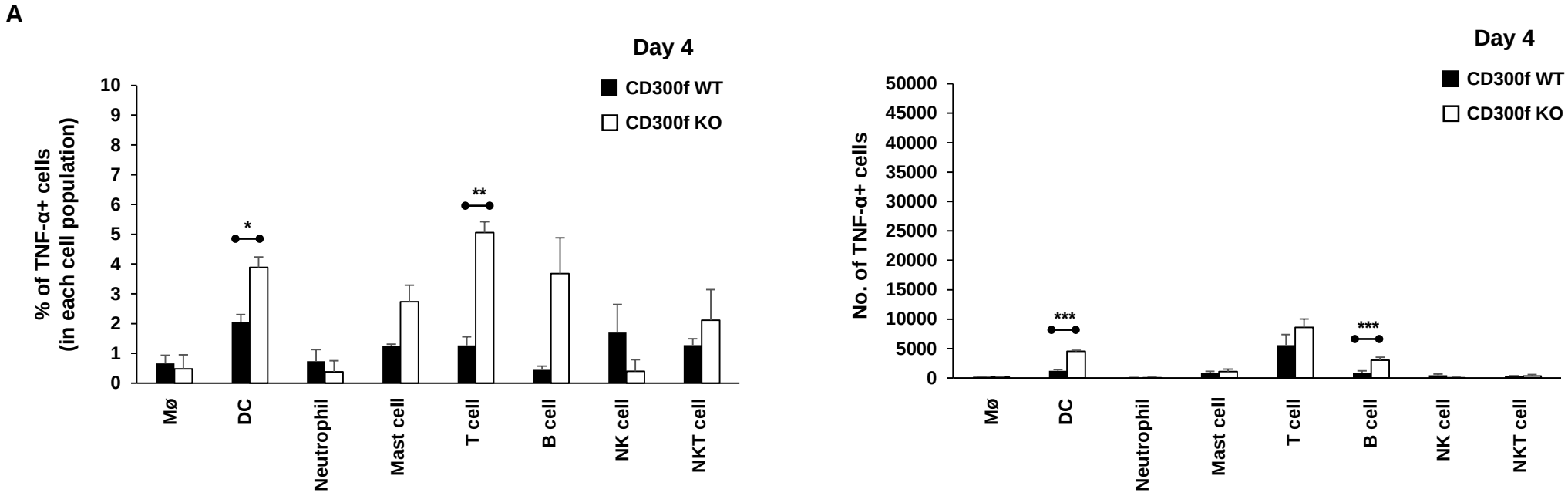
Supplemental Figure 1: Gut microbiota is not responsible for the enhanced sensitivity to colonic inflammation in *Cd300f*$^{-/-}$ mice. (**A**) Phylum-level composition of intestinal bacteria in the indicated mice. (**B-E**) *Cd300f*$^{+/+}$ and *Cd300f*$^{-/-}$ mice were co-housed for 2 weeks, and then given drinking water containing 2.5% DSS for 7 days, followed by unadulterated drinking water for another 2 days. DAI was determined during and after DSS administration (**B**). On day 9, the colon length (**C**), macroscopic inflammation score (**D**) and microscopic inflammation score (**E**) were determined. Images in **E** show H&E staining of colon tissues; scale bar 500 μm. Data are expressed as means + S.E.M. (n =5, each group). Two-tailed paired Student's t test was used to determine statistical significance (**p<0.01, ***p<0.001).



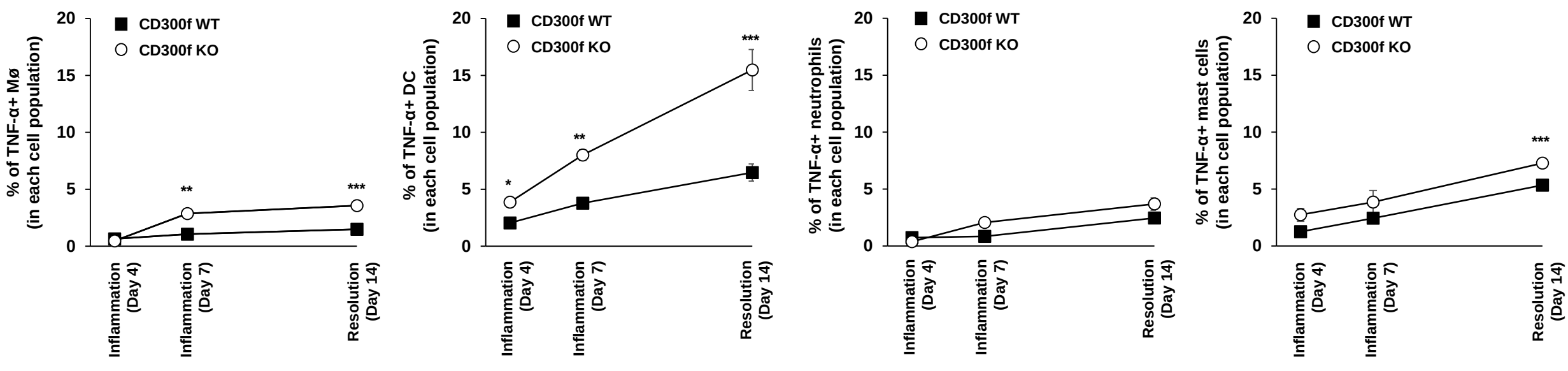
Supplemental Figure 2: Cell populations in the lamina propria of *Cd300f*$^{+/+}$ and *Cd300f* $^{-/-}$ mice. (**A** and **B**) *Cd300f*$^{+/+}$ or *Cd300f* $^{-/-}$ mice were given drinking water containing 2.5% DSS for 7 days (**A**), followed by unadulterated drinking water for another 7 days (**B**). Lamina propria immune cells were isolated from the colon of DSS-treated *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice, and the number and the percentage of total immune cells ($CD45^+$) and neutrophils ($CD45^+$ $Ly6G^+$ $CD11c^-$ $F4/80^-$) was determined by flow cytometry. Data are expressed as means + S.E.M. (n =5, each group). Two-tailed paired Student's t test was used to determine statistical significance (*p<0.05, **p<0.01, ***p<0.001).



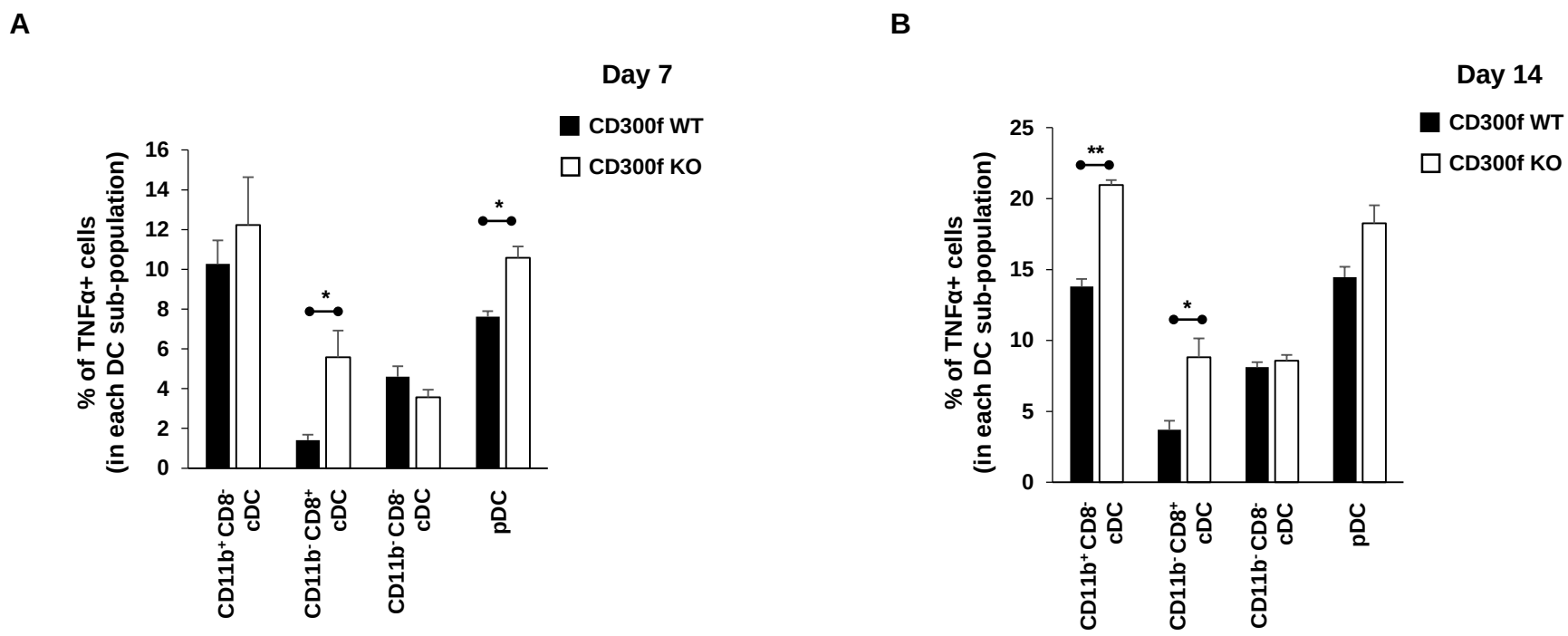
Supplemental Figure 3: Analysis of TNF-α and INF-γ signal intensities in the colon tissue. Colon sections from *Cd300f*$^{+/+}$ and *Cd300f*$^{-/-}$ mice, collected on day 7 (Inflammation) or day 14 (Resolution), were stained for TNF-α (**A**) or IFN-γ (**B**). The average pixel intensity in the collected images (n=15 in each group) was measured and is summarized in the bar graph; error bars show S.E.M. Two-tailed paired Student's t test was used to determine statistical significance (*p<0.05, ***p<0.001).



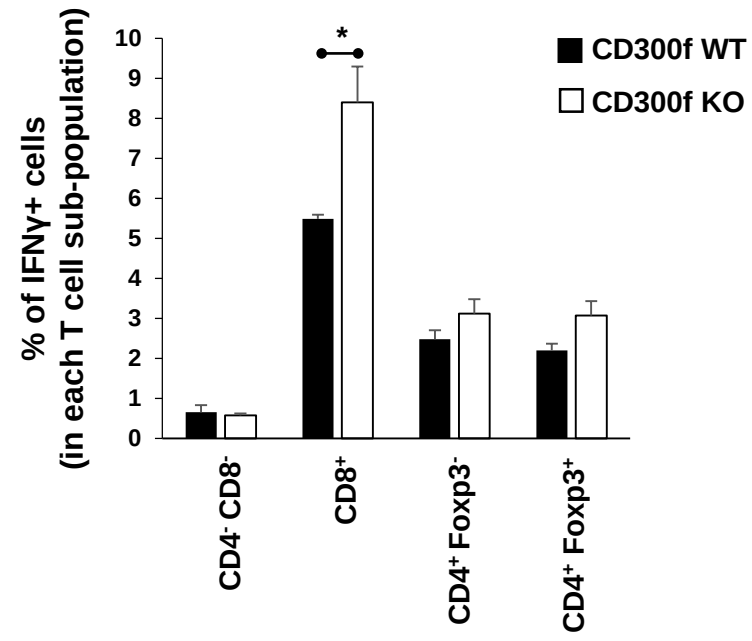
Supplemental Figure 4: Analysis of lamina propria cells producing TNF-α and IFN-γ at the early phase of colonic inflammation. (**A** and **B**) Lamina propria cells were isolated from the colons of DSS-treated *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice collected on day 4 of DSS treatment. The intracellular expression of TNF-α (**A**) and IFN-γ (**B**) was determined by flow cytometry in the following cell populations: macrophages (CD45^{+} F4/80^{+} CD11b^{+} CD14^{+} Ly6G^{-} CD11c^{-}), DC (CD45^{+} CD11c^{+} F4/80^{-} Ly6G^{-} CD64^{-}), neutrophils (CD45^{+} CD11b^{+} Ly6G^{+} CD11c^{-} F4/80^{-}), mast cells (CD45^{+} CD11b^{+} FcεRI^{+}), T cells (CD45^{+} CD3^{+}), B cells (CD45^{+} CD3^{-} CD19^{+}), NK cells (CD45^{+} CD3^{-} NK1.1^{+}), and NKT cells (CD45^{+} CD3^{+} NK1.1^{+}). The graphs show the percentages (*left*) and total number (*right*) of cells expressing TNF-α (**A**), or IFN-γ (**B**). Data are expressed as means + S.E.M. (n =3, each group). Two-tailed paired Student's t test was used to determine statistical significance (**p*<0.05, ***p*<0.01, ****p*<0.001).



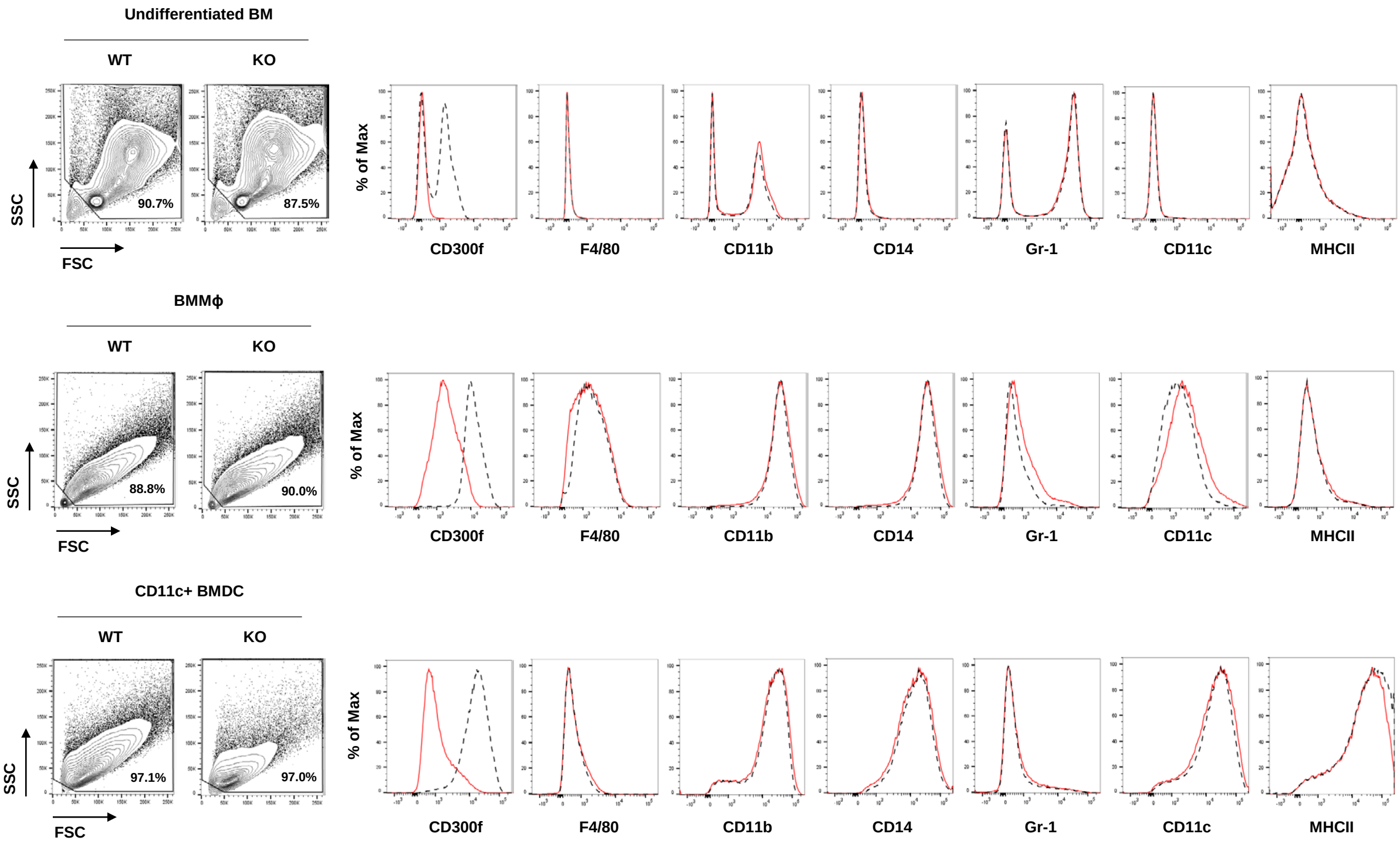
Supplemental Figure 5: Analysis of TNF-α-producing cells in the lamina propria during gut inflammation. Lamina propria cells were isolated from the colons of DSS-treated *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice collected on day 4, 7, and 14. The intracellular expression of TNF-α was determined by flow cytometry in the following cell populations: macrophages (CD45^{+} F4/80^{+} CD11b^{+} CD14^{+} Ly6G^{-} CD11c^{-}), DC (CD45^{+} CD11c^{+} F4/80^{-} Ly6G^{-} CD64^{-}), neutrophils (CD45^{+} CD11b^{+} Ly6G^{+} CD11c^{-} F4/80^{-}), mast cells (CD45^{+} CD11b^{+} FcεRI^{+}), T cells (CD45^{+} CD3^{+}), B cells (CD45^{+} CD3^{-} CD19^{+}), NK cells (CD45^{+} CD3^{-} NK1.1^{+}), and NKT cells (CD45^{+} CD3^{+} NK1.1^{+}). The graphs summarize the data from Figure 3 and Supplemental Figure 10, and illustrate the percentages of cells expressing TNF-α at the indicated times. Data are expressed as means ± S.E.M. (n = 3-8, each group). Two-tailed paired Student's t test was used to determine statistical significance (*p<0.05, **p<0.01, ***p<0.001).



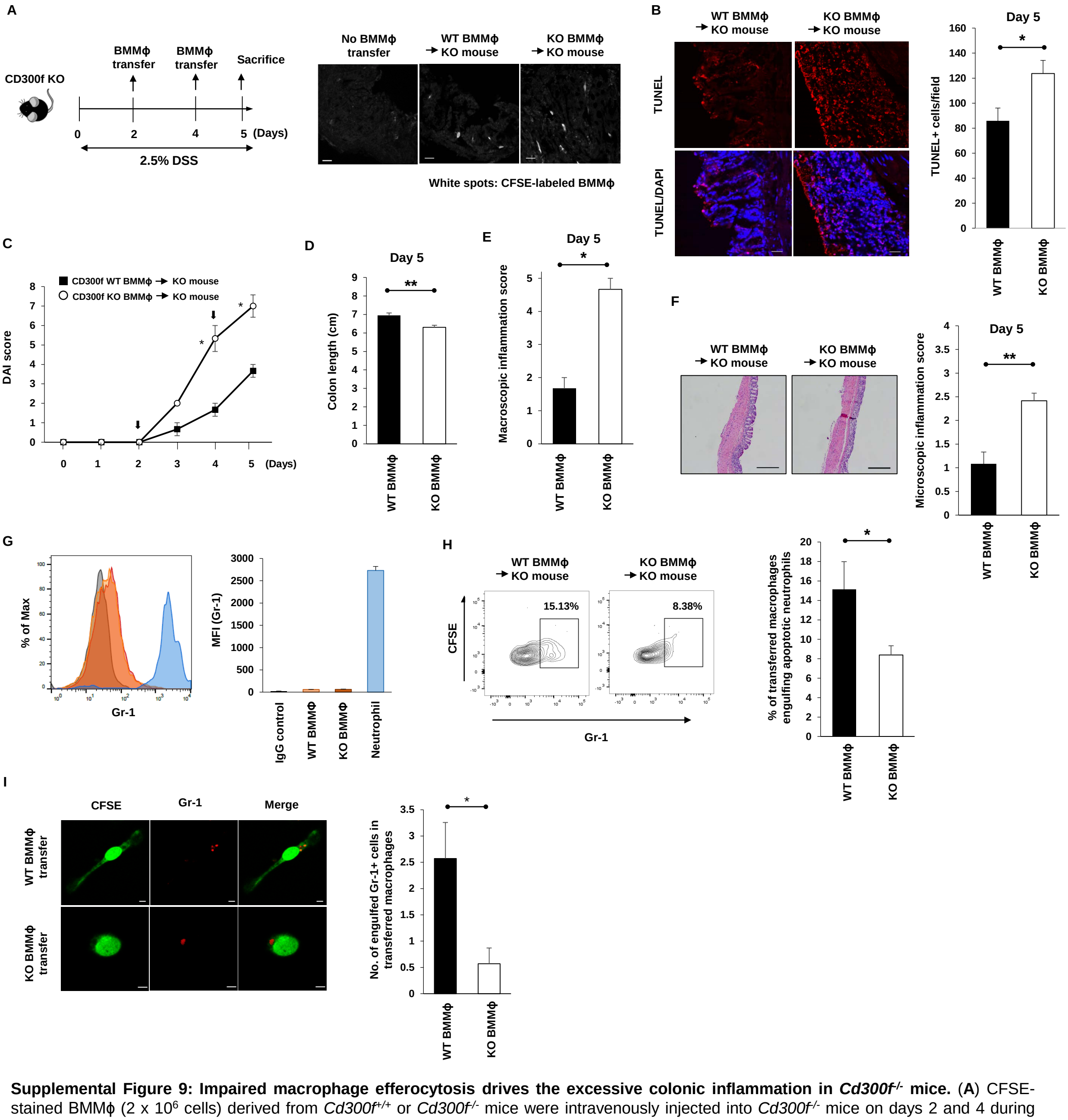
Supplemental Figure 6: Analysis of TNF-α production by DC sub-populations in the lamina propria. Lamina propria cells were isolated from the colon of DSS-treated *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice on day 7 (**A**) or day 14 (**B**) after 2.5% DSS treatment. The intracellular levels of TNF-α in DC cell sub-populations including myeloid CD11b^{+} CD8^{-}, CD11b^{-} CD8^{+}, CD11b^{-} CD8^{-} DC, and plasmacytoid DC (pDC) were determined by flow cytometry. The graph shows the percentage of cells expressing TNF-α, as means + S.E.M. (n =3, each group). Two-tailed paired Student's t test was used to determine statistical significance (*p<0.05, **p<0.01).



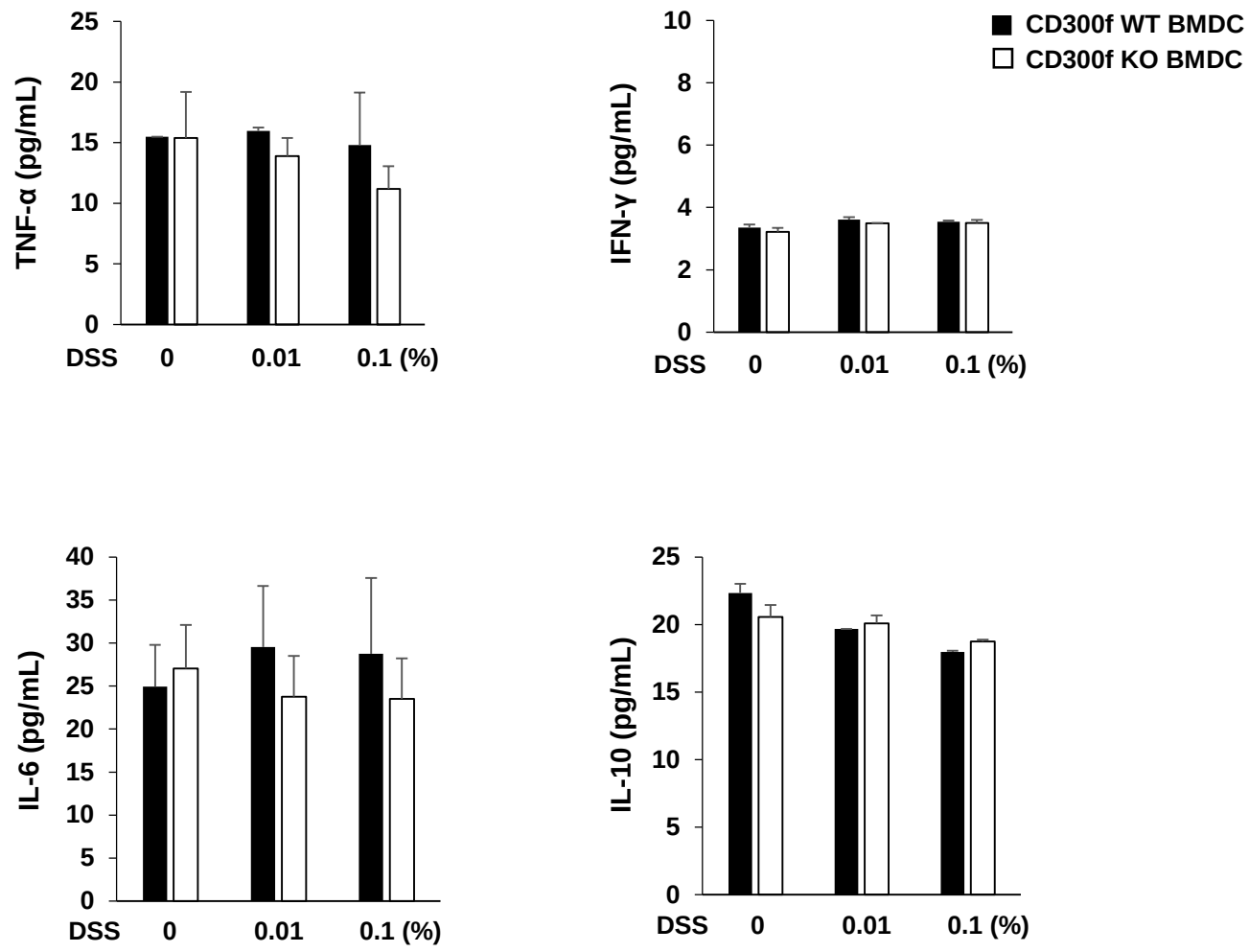
Supplemental Figure 7: CD8$^+$ T cells constitute the major T cell population producing IFN-γ in the lamina propria of *Cd300f*$^{-/-}$ mice. Lamina propria cells were isolated from the colon of DSS-treated *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice on day 14 after 2.5% DSS treatment. The intracellular levels of IFN-γ in T cell sub-populations including CD4$^-$ CD8$^-$, CD8$^+$, CD4$^+$ FoxP3$^-$, and CD4$^+$ FoxP3$^+$ T cells were determined by flow cytometry. The graph shows the percentage of cells expressing IFN-γ, as means + S.E.M. (n =5, each group). Two-tailed paired Student's t test was used to determine statistical significance (*p<0.05).



Supplemental Figure 8: Analysis of BMMϕ and BMDC purity. Bone marrow cells (BM) were isolated from *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice and differentiated to macrophages (BMMϕ) or DC (BMDC) as described in Methods section. BMMϕ were crudely purified by removal of non-adherent cells, and BMDC were purified from non-adherent cells using CD11c selection (see Methods for details). Cell identity and population purity were analyzed by flow cytometry. Dot plots show the forward scatter (FSC) and side scatter (SSC) distribution of analyzed cells, and the gating strategy. Histograms illustrate the indicated marker distribution on the surface of cells derived from *Cd300f*$^{+/+}$ (WT; black dashed lines) or *Cd300f*$^{-/-}$ mice (KO; red solid lines). Results are representative of two independent experiments.

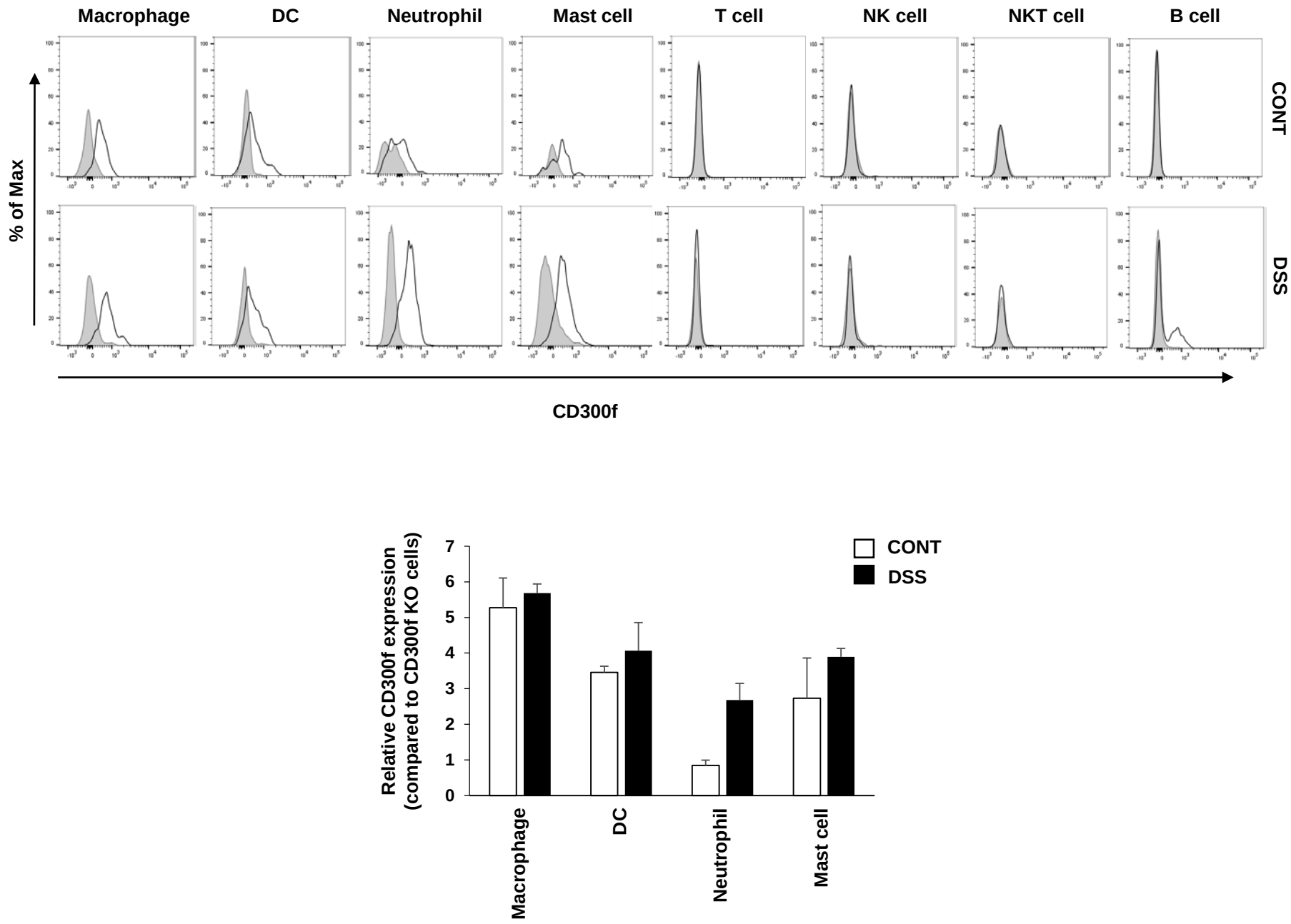


Supplemental Figure 9: Impaired macrophage efferocytosis drives the excessive colonic inflammation in *Cd300f*$^{-/-}$ mice. (**A**) CFSE-stained BMMϕ (2 x 10^6 cells) derived from *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice were intravenously injected into *Cd300f*$^{-/-}$ mice on days 2 and 4 during DSS administration. The presence of the transferred BMMϕ (white spots) in the colon tissues was verified by confocal microscopy (scale bars: 20 µm). (**B**) Representative immunofluorescence staining for TUNEL (red) in frozen colon sections from DSS-treated *Cd300f*$^{-/-}$ mice injected with BMMϕ derived from *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice. Nuclei were stained with DAPI (blue). Scale bars: 20 µm. The graph shows the quantification of TUNEL$^+$ cells per field of view (3 fields per colon section). (**C**) DAI scored during DSS administration; the arrows indicate the time of BMMϕ transfer. (**D-F**) On day 5, the colon length (**D**), macroscopic inflammation score (**E**) and microscopic inflammation score (**F**) were determined. The pictures in **F** illustrate representative images of the colon in the indicated mice (H&E-staining, scale bars: 500 µm). (**G**) Cell surface expression of Gr-1 on BMMϕ derived from *Cd300f*$^{+/+}$ and *Cd300f*$^{-/-}$ mice, or neutrophils. The histograms show a representative flow cytometry result. The bar graph shows the quantification of Gr-1 mean fluorescence intensity (MFI) values. (**H** and **I**) Spleens were collected from DSS-treated *Cd300f*$^{-/-}$ mice i.v. injected with CFSE-labeled BMMϕ derived from *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice. (**H**) The percentage of transferred BMMϕ engulfing AC (CFSE$^+$ F4/80$^+$ Gr-1$^+$) in the spleen was determined using flow cytometry. (**I**) Images show representative examples of transferred BMMϕ (CFSE$^+$; green) engulfing apoptotic neutrophils (Gr-1$^+$; red); scale bars: 5 µm. Data are expressed as means + S.E.M (n = 12, each group in **B**, n = 4, each group in **D**-**F**, n = 4 in **H**, n = 10 in **I**). Two-tailed paired Student's t test was used to determine statistical significance (*p<0.05, **p<0.01).

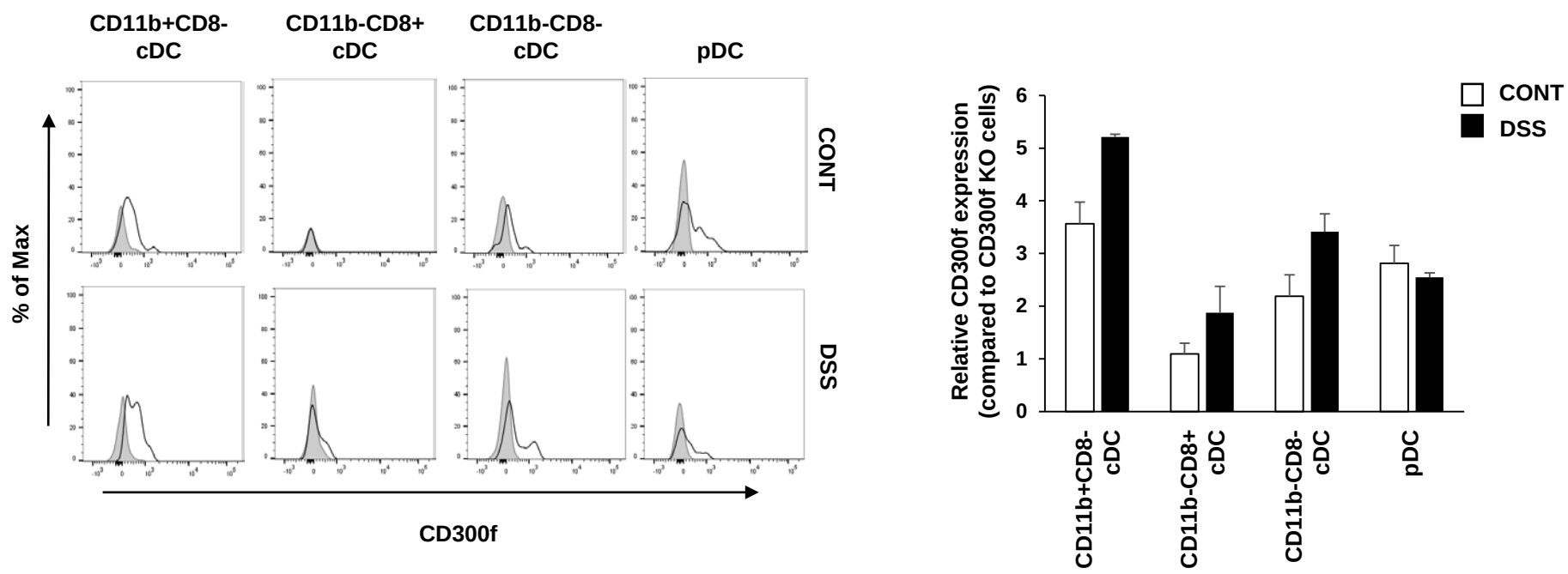


Supplemental Figure 10: DSS alone does not alter cytokine production by *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ BMDC. Purified CD11c^{+} BMDC from *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice were treated with 0.01 or 0.1% DSS for 24 h. The cell culture media were collected, and the levels of the indicated cytokines were determined. Data are expressed as means + S.E.M. from 3 separate experiments.

(A)



(B)



Supplementary figure 11. CD300f expression on lamina propria cells. Lamina propria cells were isolated from the colon of *Cd300f*$^{+/+}$ or *Cd300f*$^{-/-}$ mice treated without or with DSS on day 7. (**A**) Surface expression of CD300f on colonic macrophages (CD45^{+} F4/80^{+} CD11b^{+} CD14^{+} Ly6G^{-} CD11c^{-}), DC (CD45^{+} CD11c^{+} F4/80^{-} Ly6G^{-} CD64^{-}), neutrophils (CD45^{+} CD11b^{+} Ly6G^{+} CD11c^{-} F4/80^{-}), mast cells (CD45^{+} CD11b^{+} FcεRI^{+}), T cells (CD45^{+} CD3^{+}), NK cells (CD45^{+} CD3^{-} NK1.1^{+}), and NKT cells (CD45^{+} CD3^{+} NK1.1^{+}) was determined by flow cytometry. (**B**) Surface expression of CD300f on DC sub-populations was determined by flow cytometry. Histograms in (**A**) and (**B**) illustrate the expression level of CD300f on the surface of cells derived from *Cd300f*$^{+/+}$ (black solid line) or *Cd300f*$^{-/-}$ mice (gray histograms). The graph shows the fold changes in CD300f expression on cells derived from *Cd300f*$^{+/+}$ mice compared to cells derived from *Cd300f*$^{-/-}$ mice. Results are representative of three independent experiments.