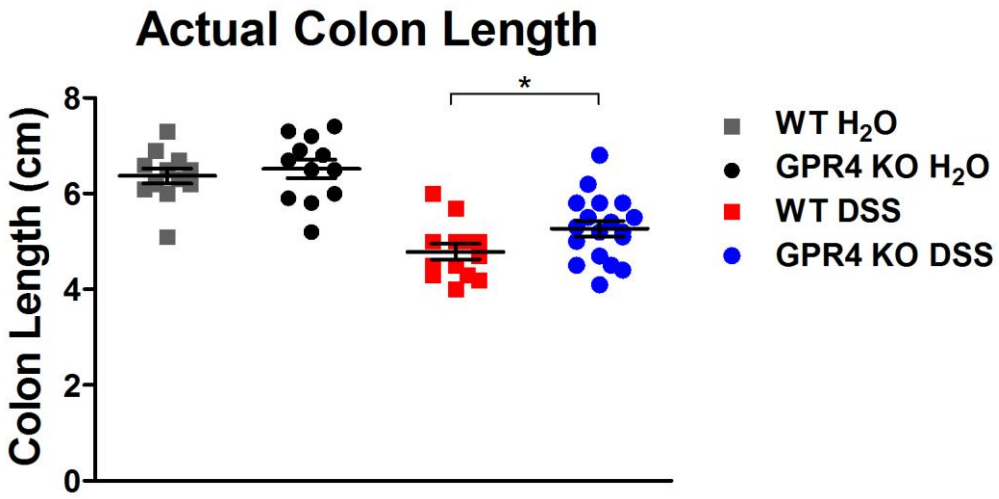
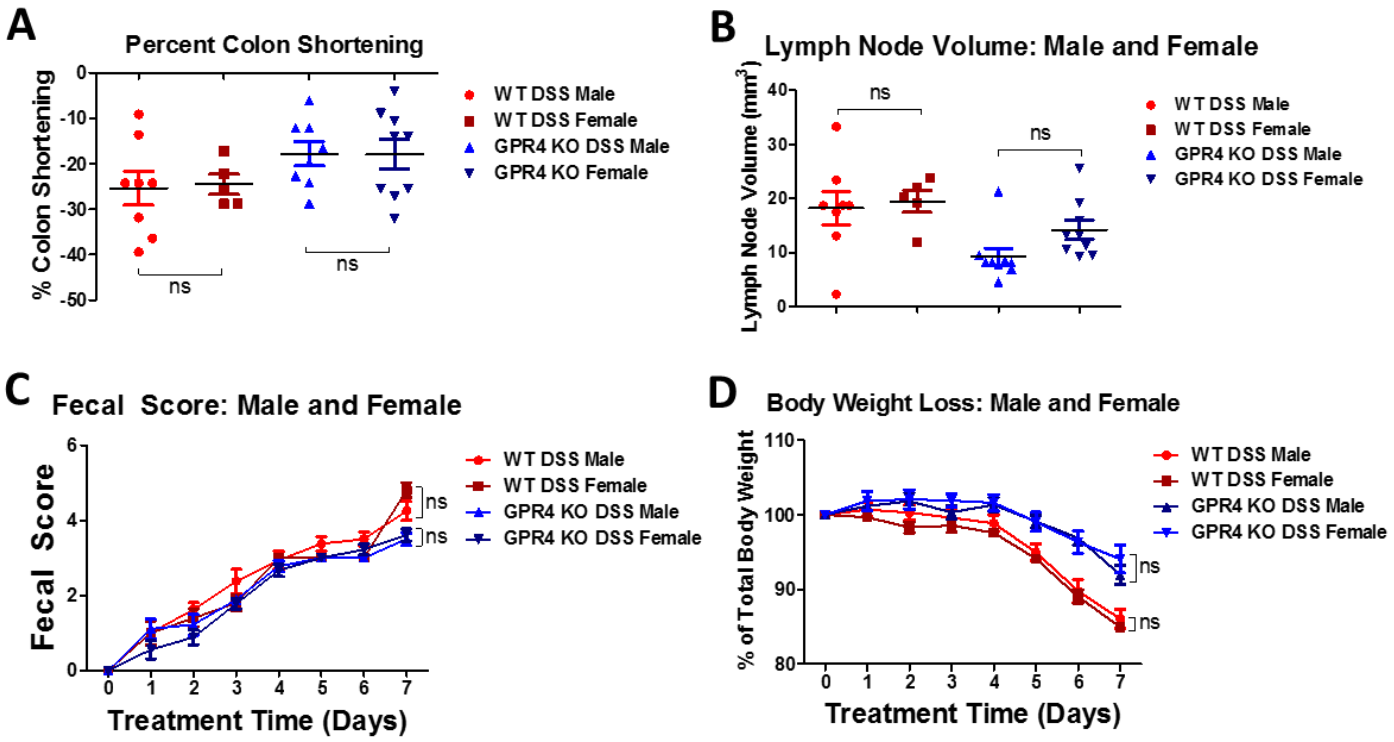
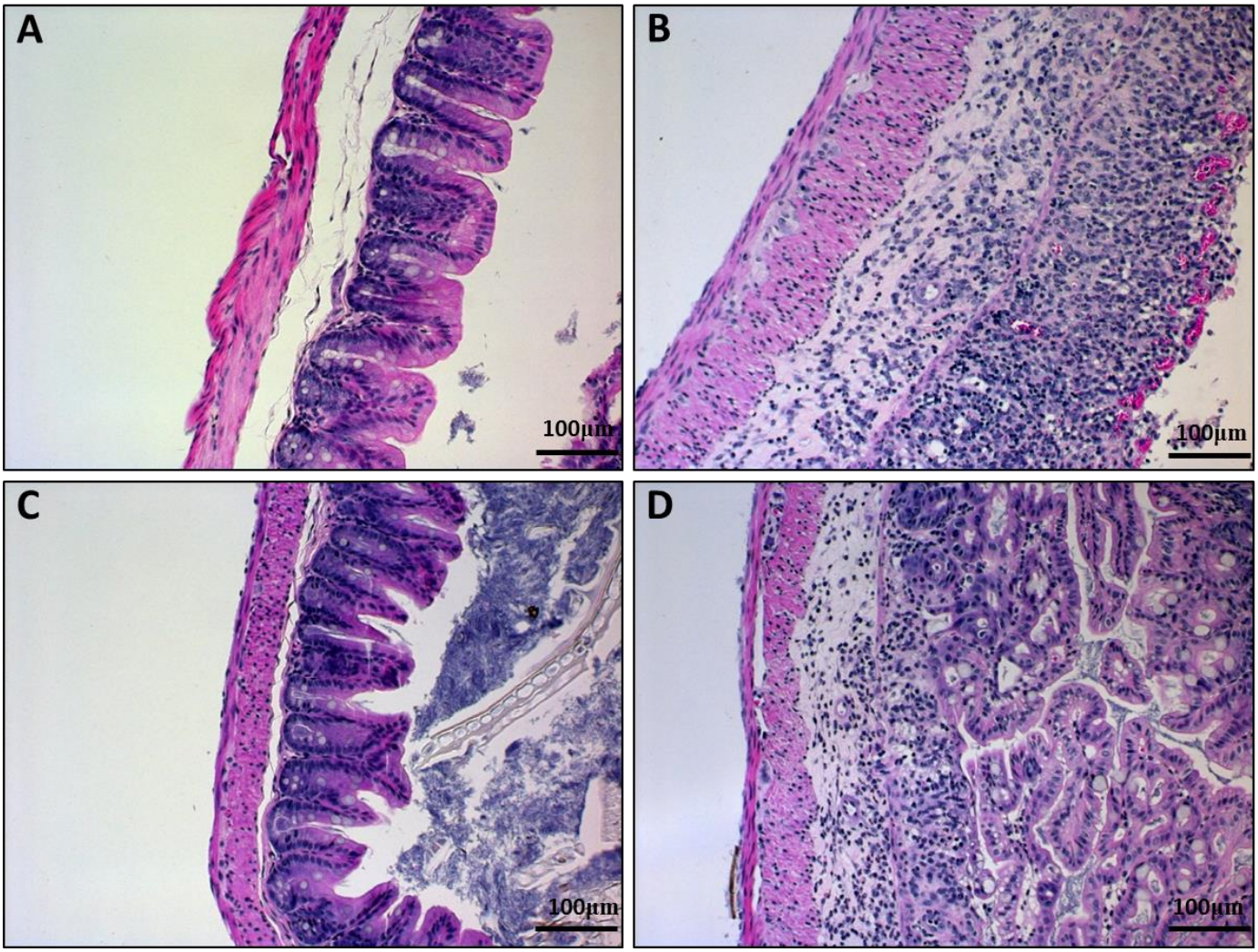


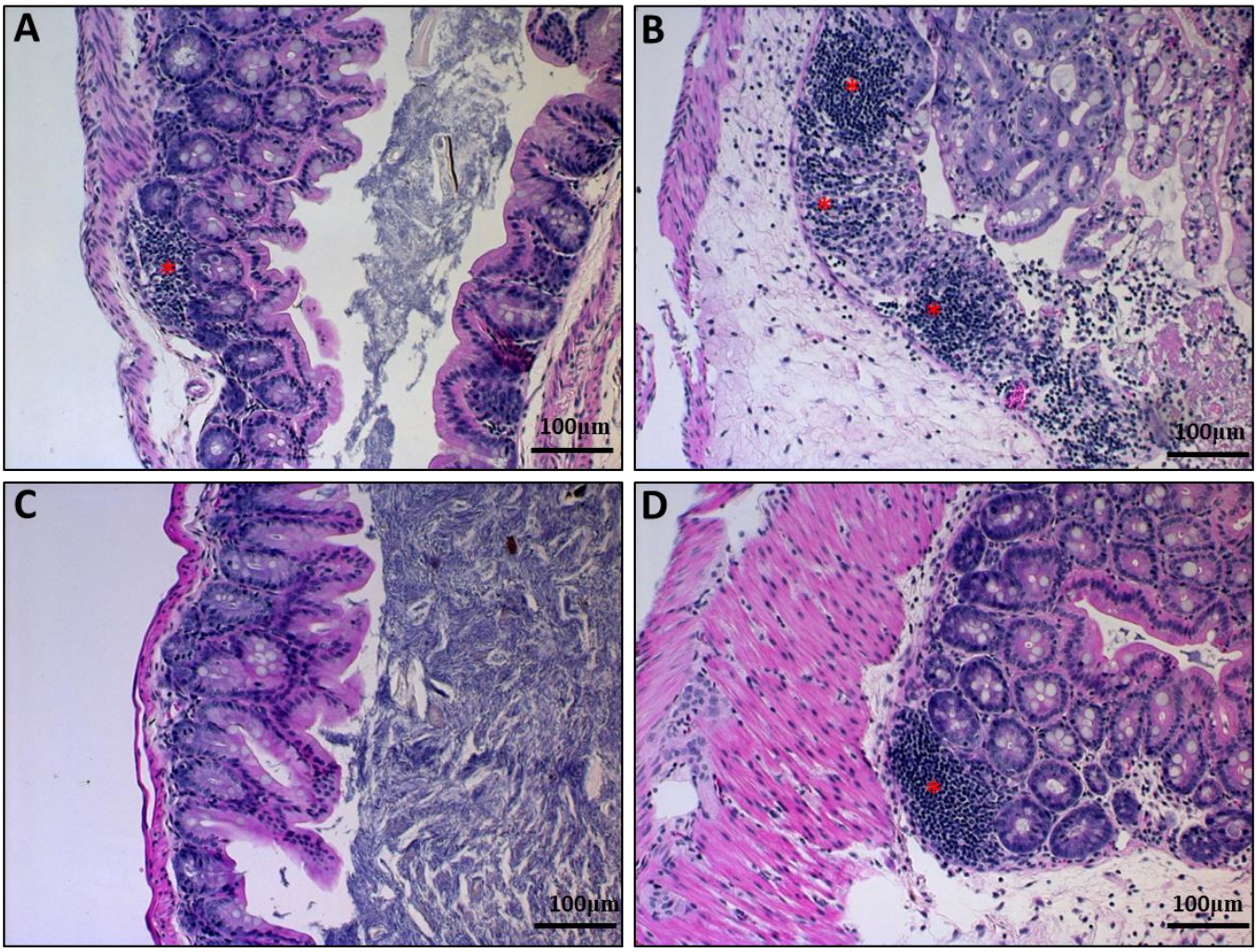
Supplementary Fig. S1. Actual colon length as a clinical parameter of intestinal inflammation. Colon lengths of WT control (n=12), WT-DSS (n=13), GPR4 KO control (n=12), and GPR4 KO-DSS (n=18) are presented in centimeters. Data are presented as mean ± SEM and analyzed for statistical significance using the unpaired *t*-test between two groups indicated on graph. (*$P < 0.05$)



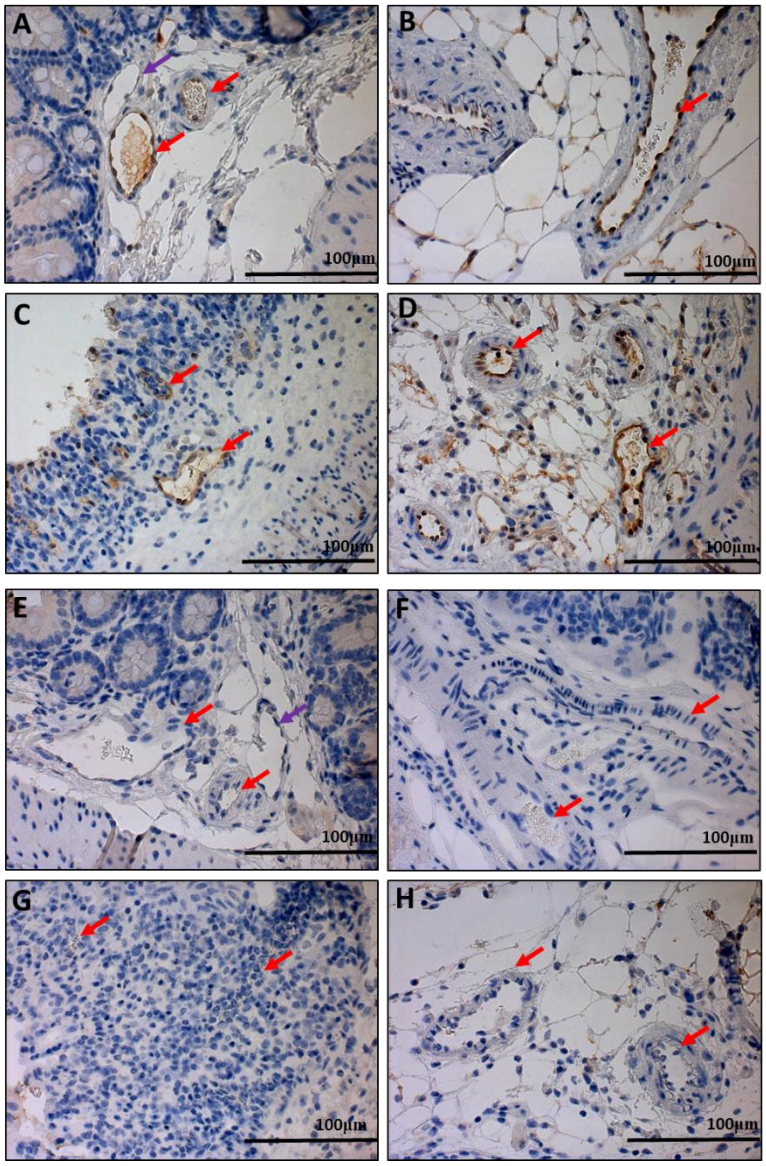
Supplementary Fig. S2. Clinical phenotype and macroscopic disease indicators. (A) Percent colon shortening male vs. female mice, (B) mesenteric lymph node volume male vs. female mice, (C) fecal score male vs. female mice, and (D) percent body weight loss male vs. female mice. Data are presented as mean ± SEM and analyzed for statistical significance using the unpaired *t*-test between two groups indicated on graph. *ns*: not significant.



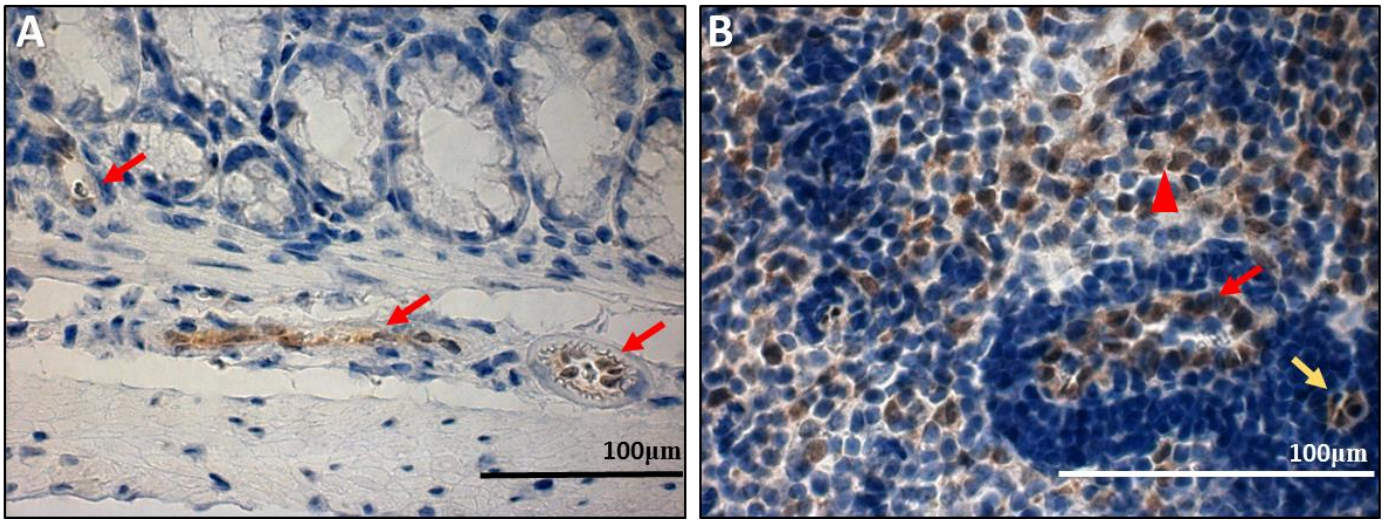
Supplementary Fig. S3. H&E representative pictures of cecum in (A) WT control, (B) WT-DSS, (C) GPR4 KO control, and (D) GPR4 KO-DSS. 20× microscope objective.



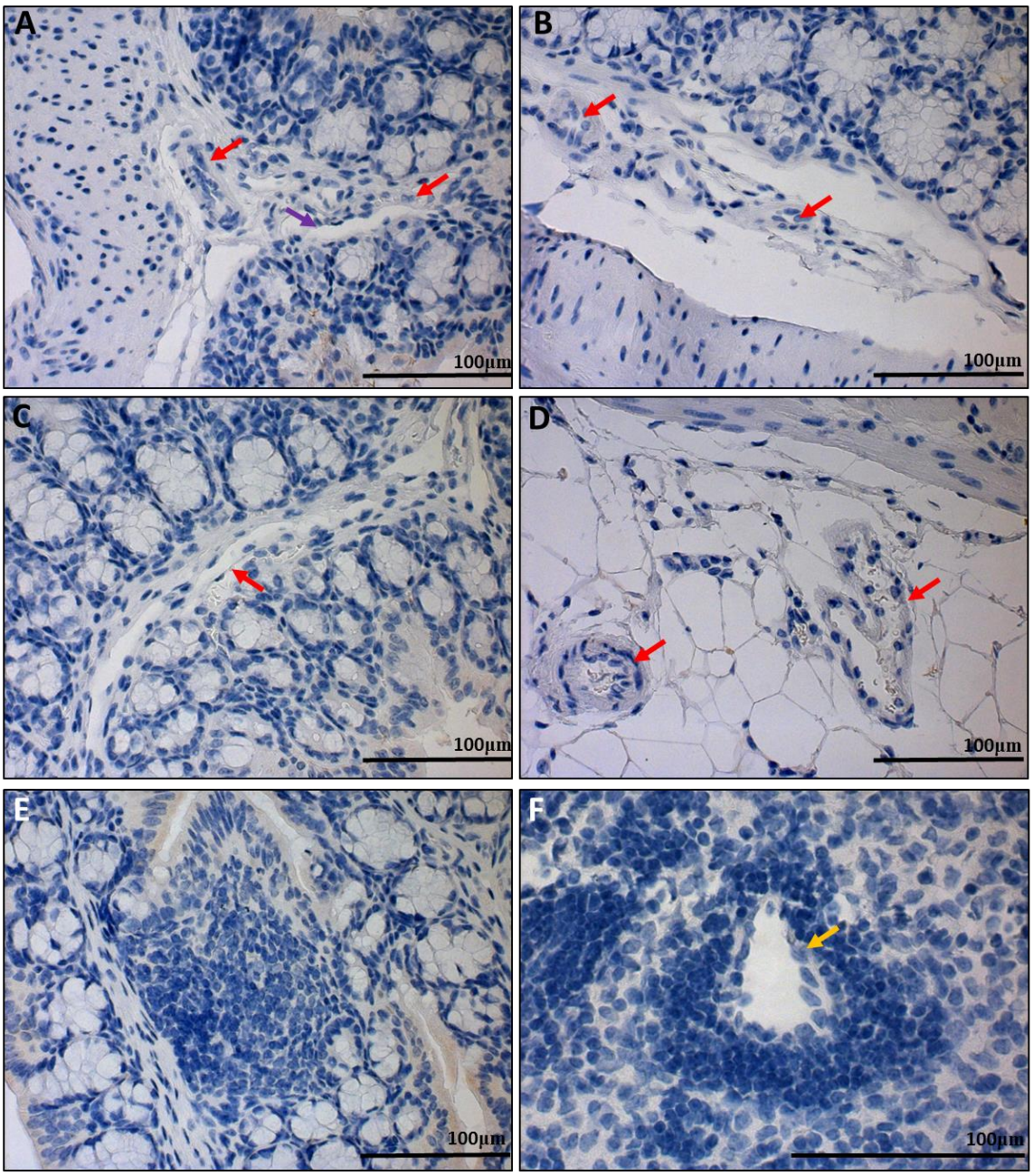
Supplementary Fig. S4. H&E representative pictures of ILFs in cecum. (A) WT control, (B) WT-DSS, (C) GPR4 KO control, and (D) GPR4 KO-DSS. 20× microscope objective. Red asterisk indicates ILF.



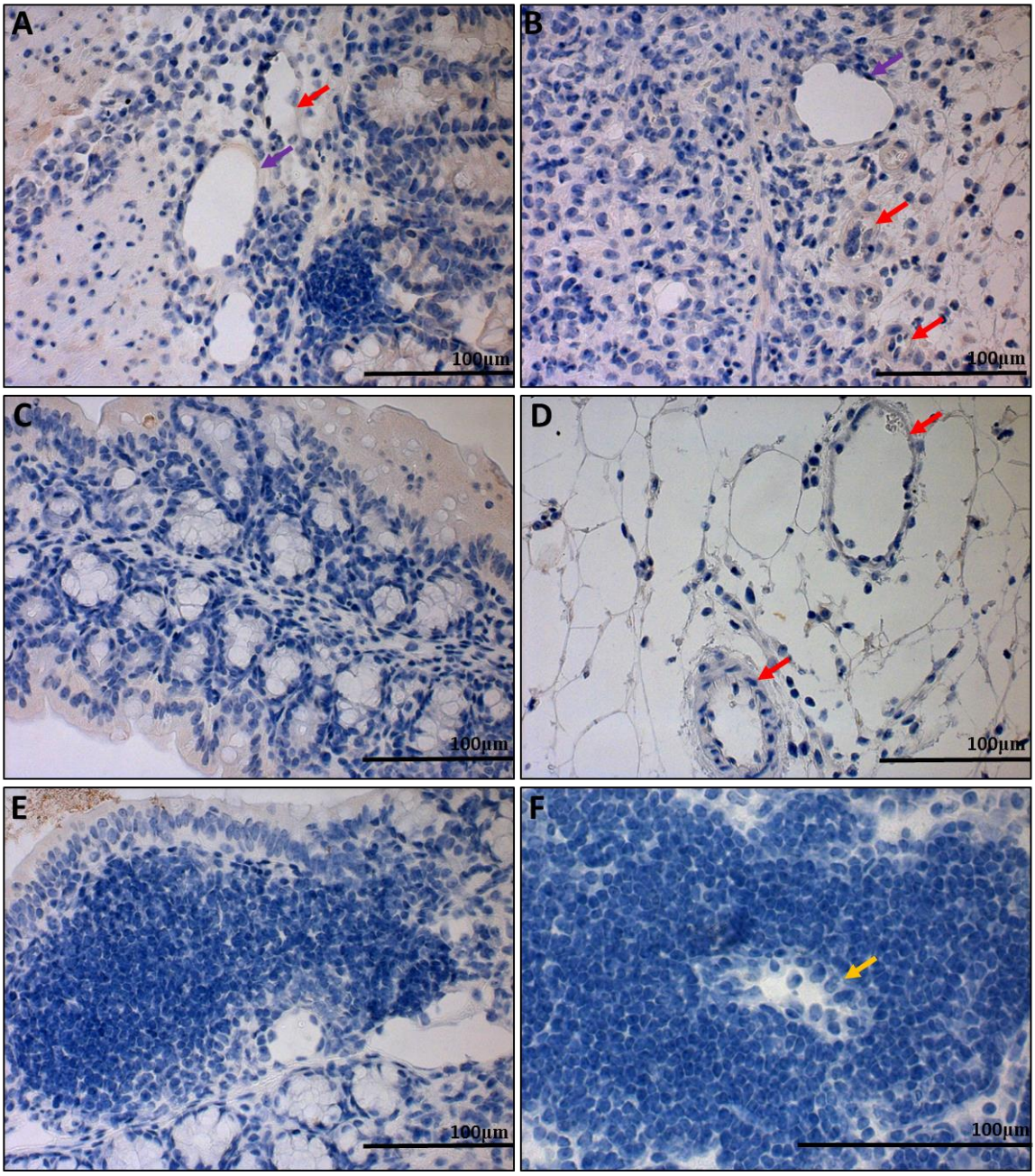
Supplementary Fig. S5. GFP knock-in as a surrogate marker for GPR4 expression in GPR4 KO mouse cecum. Immunolabeling of GFP was performed in GPR4 KO cecum tissue. Similarly to mouse colon tissue, GFP expression could be visualized in the intestinal microvascular endothelial cells, ex-mural blood vessels, arteries in both control and inflamed cecum tissues. Lymphatics had very low GFP expression. (A) GPR4 KO untreated cecum submucosa blood vessels and (B) ex-mural vessels compared to (C) inflamed GPR4 KO-DSS submucosa blood vessels and (D) ex-mural vessels. No GFP signal could be detected in (E-F) WT control untreated cecum tissues and (G-H) WT-DSS cecum tissues. 40× microscope objective. Red arrows indicate blood vessels and purple arrows indicate lymphatics.



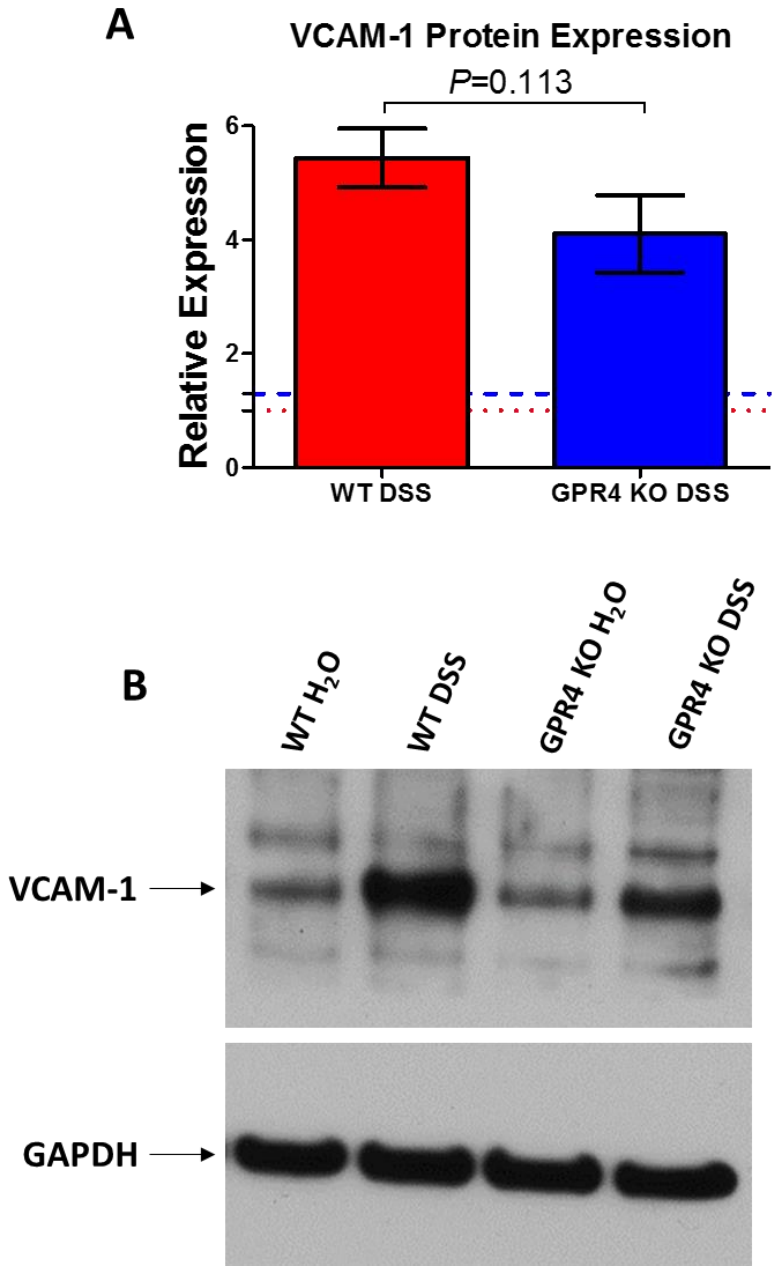
Supplementary Fig. S6. GFP immunohistochemistry of GPR4 heterozygous mouse colon and mesenteric lymph node. GFP expression can be observed in the same cell types observed in GPR4 KO homozygous mouse tissues. (A) GFP can be detected in colon blood vessels. 40× microscope objective. (B) GFP expression in the lymph node can be observed in resident histiocytes, blood vessels, and HEVs. 63× microscope objective. Red arrow heads indicate macrophages, red arrows indicate blood vessels, and yellow arrows indicate HEVs.



Supplementary Fig. S7. GFP immunohistochemistry of WT control colon. No visible GFP expression can be detected in tissues. Minor background staining could be observed on epithelium. (A-B) Submucosa, (C) transverse folds, (D) ex-mural, (E) isolated lymphoid follicles, (F) mesenteric lymph node HEV and resident macrophages. (A-E) 40× and (F) 63× microscope objectives. Red arrows indicate blood vessels, yellow arrows indicate HEVs, and purple arrows indicate lymphatics.



Supplementary Fig. S8. GFP immunohistochemistry of WT-DSS treated colon. No visible GFP expression can be detected in tissues. Minor background staining could be observed on epithelium, luminal content, and connective tissues. (A-B) Submucosa, (C) transverse folds, (D) ex-mural, (E) isolated lymphoid follicles, and (F) mesenteric lymph node HEV and resident macrophages. (A-E) 40× and (F) 63× microscope objectives. Red arrows indicate blood vessels, yellow arrows indicate HEVs, and purple arrows indicate lymphatics.



Supplementary Fig. S9. Western blot analysis of VCAM-1 protein expression in mouse colon tissues. VCAM-1 protein expression was reduced in GPR4 KO-DSS (n=9) colon tissues when compared to WT-DSS (n=9), although not statistically significant (*P*=0.113). Target bands are indicated by an arrow. Red and blue dotted lines indicate WT-control (n=9) and GPR4 KO control (n=9) quantification, respectively. Data are presented as mean ± SEM and analyzed for statistical significance using the unpaired t-test between WT-DSS and GPR4 KO-DSS mice.