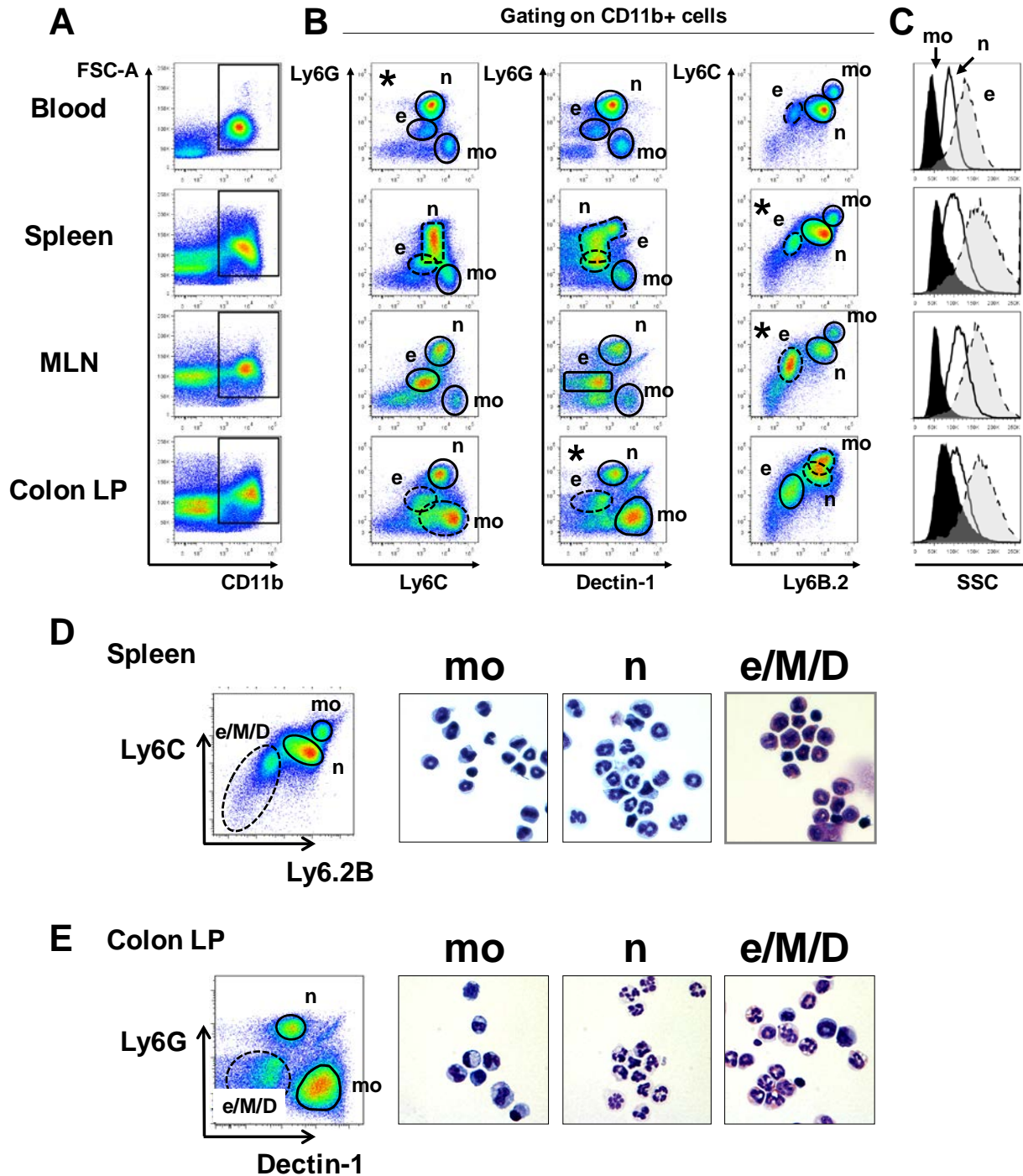
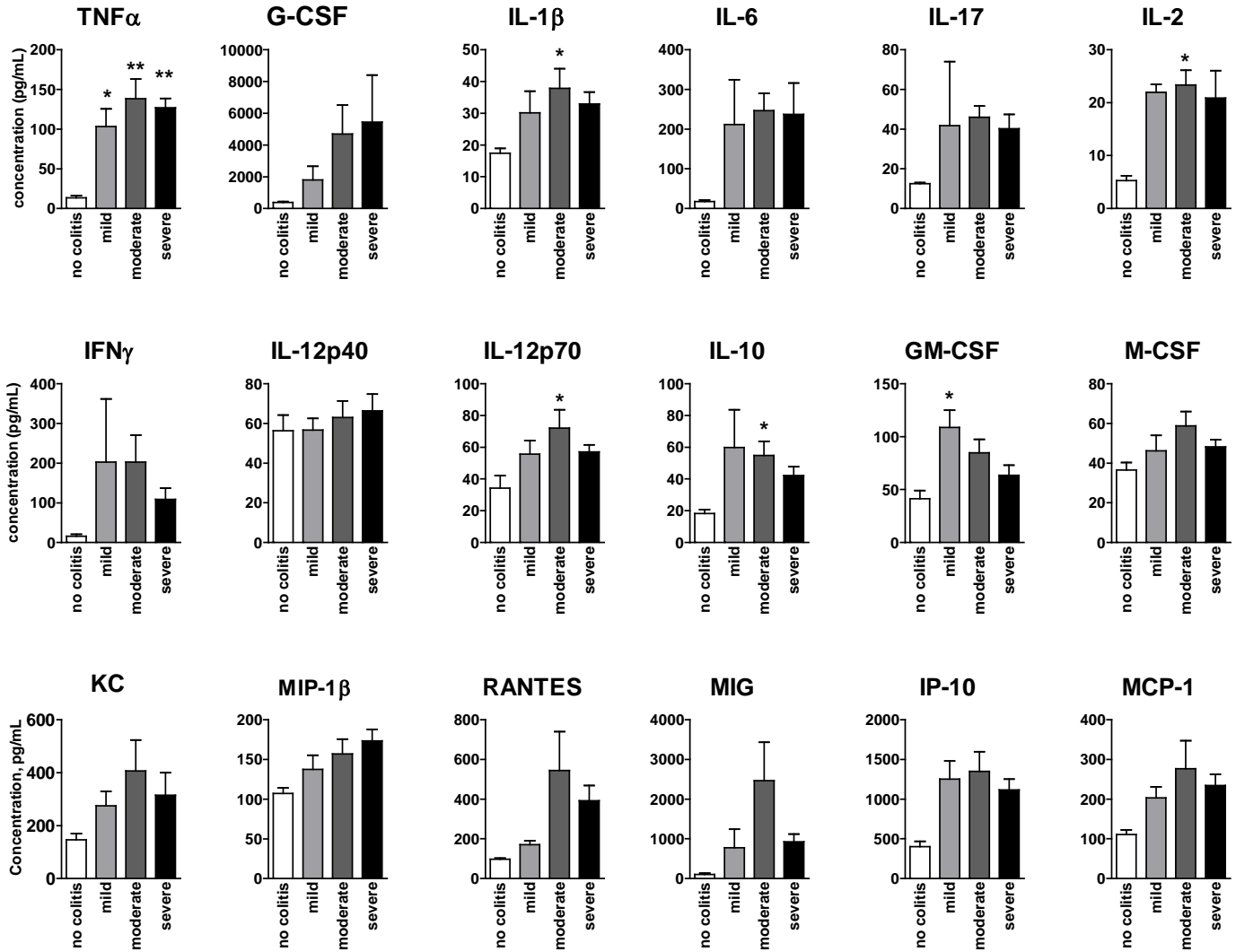


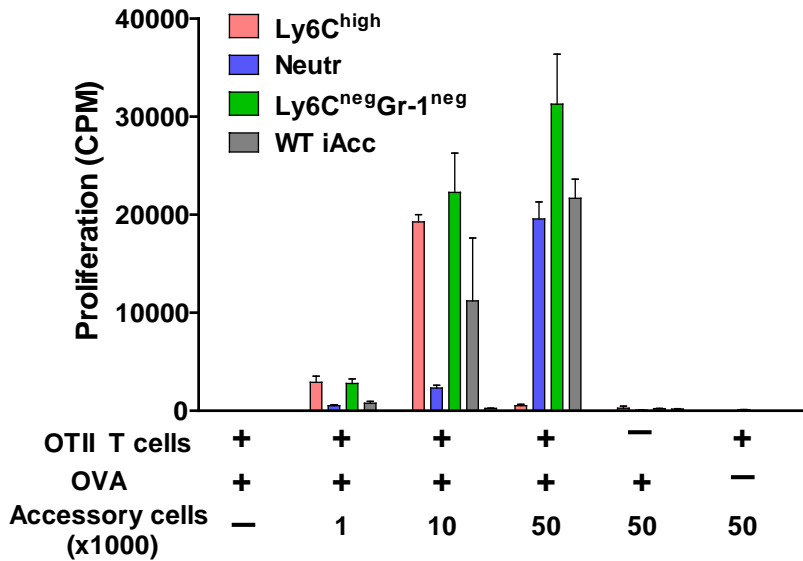
Supplementary figure 1. Identification of myeloid cells in the indicated tissues of colitic mice using surface antibodies. A. Mac-1+ cells were initially gated to exclude doublet populations and were further analyzed using the indicated markers. Neutrophils (n), monocytes (mo), eosinophils (e), and macrophages/DC (M/D) are marked next to the circled populations on the representative dot plots. Dotted lines on some plots indicate approximate location of specified populations of cells. Asterisks indicate gating strategy that were used to obtain side scatter (SSC) histogram profiles (right) to show differences in granularity. Cytospin images of indicated populations isolated from spleen (B) and cLP (C). Populations shown on the dot plots on the left were sorted and cells were analyzed by cytopsin following by Diff-Quik staining to reveal cellular morphology. Ly6C^{low/neg}Ly6.2B^{neg} cells in addition to eosinophils contain macrophages and DCs (M/D). Dotted lines on some plots indicate approximate location of specified populations of cells.



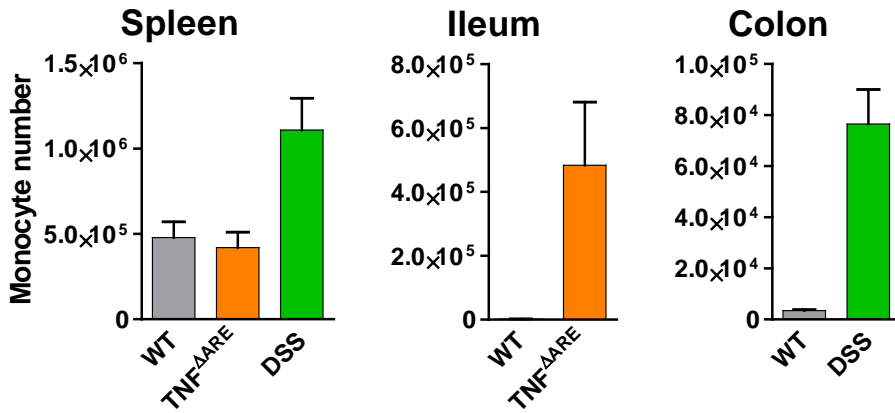
Supplementary figure 2. Plasma levels of cytokine and chemokines in RAG-1^{-/-} mice that developed disease of varying severity following adoptive transfer of CD45RB^{high} T cells, based on their blinded histopathological scores. No colitis group corresponded to scores of 0-2, mild colitis – 3-7, moderate – 8-12, and severe–13-17. Significant differences from non-colitic mice (e.g. RAG-1^{-/-} mice injected with RB^{high} T cells but did not develop colitis) are indicated by*, where * indicates p<0.05, ** indicates p<0.01, *** indicates p<0.001. Combined data from 3-5 mice per group.



Supplementary figure 3. Colon LP Ly6C^{high} cells directly suppress proliferation of CD4 T cells. Co-cultures were set up as described in the Materials and Methods section with the sorted colon LP CD11b+Ly6G^{neg}Ly6C^{high} cells, CD11b+Ly6G+Ly6C^{int} neutrophil-like cells, CD11b+Ly6G^{neg}Ly6C^{low/neg}, or irradiated splenocytes isolated from wild-type mice as APCs. CD4 OT2 T cells were flow-purified from spleens of OT2 mice by sorting for (CD8, CD11c, B220, CD11b)^{neg} CD4⁺ cells.

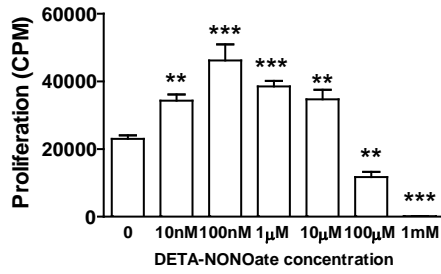


Supplementary figure 4. Acute colitis and chronic ileitis is accompanied by accumulation of inflammatory monocyte-like cells in the affected tissues and spleen. Acute colitis was induced in WT by administration of 3% dextrane sodium sulfate (DSS) in drinking water for 7 days. Heterozygous TNF^{ΔARE} and control WT mice were sacrificed at 16 weeks of age. Cells from spleen, colon, and 10cm of distal ileum were isolated and stained with fluorescently-tagged monoclonal antibodies followed by flow cytometry analysis and quantification as described in Materials and Methods.



Supplementary figure 5. Low NO levels promoted while high NO levels inhibited T cell proliferation by induction of apoptosis. CD4+ T cells were cultured on CD3-coated plates with soluble CD28 was added (1ug/ml final). A. Proliferation was assessed after 72-hours using standard methods as described in the Materials and Methods section. B. Viability was assessed after 48 hours by staining with anti-annexinV antibody and propidium iodide (PI) per manufacturer's instructions. Significant differences from T cells cultured without DETA-NONOate are indicated by *, where ** indicates $p < 0.01$, *** indicates $p < 0.001$.

A



B

