SUPPLEMENTAL MATERIAL

Kleinschek et al., http://www.jem.org/cgi/content/full/jem.20081712/DC1

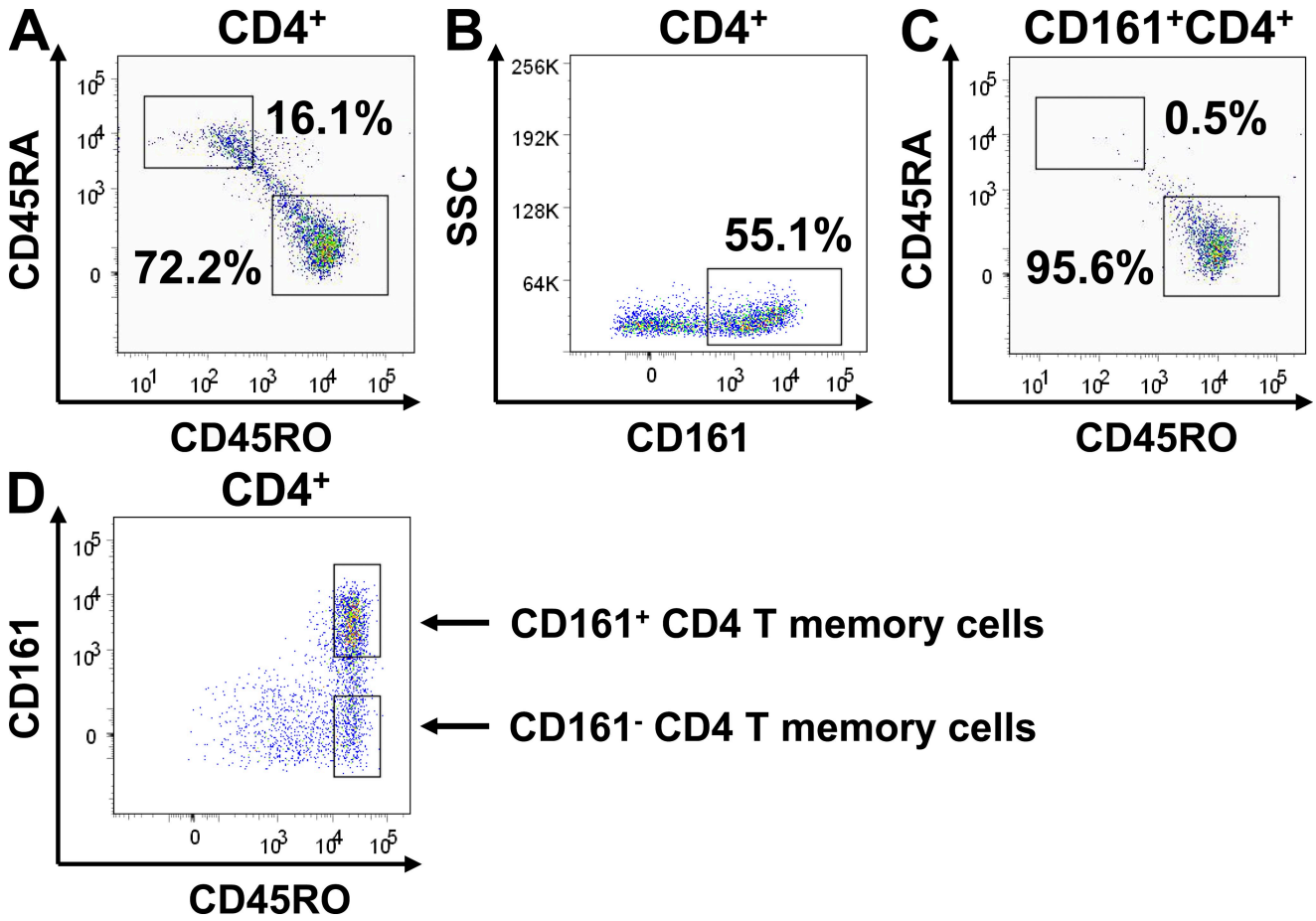


Figure S1. CD161⁺ CD4 T cells comprise a major Th cell subset in the colonic lamina propria. LPMCs were isolated from control colon specimens and were stained for CD4, CD45RA, CD45RO, and CD161. Cells are gated on lymphocytes and the markers indicated above the plots. Data are representative of at least five donors.

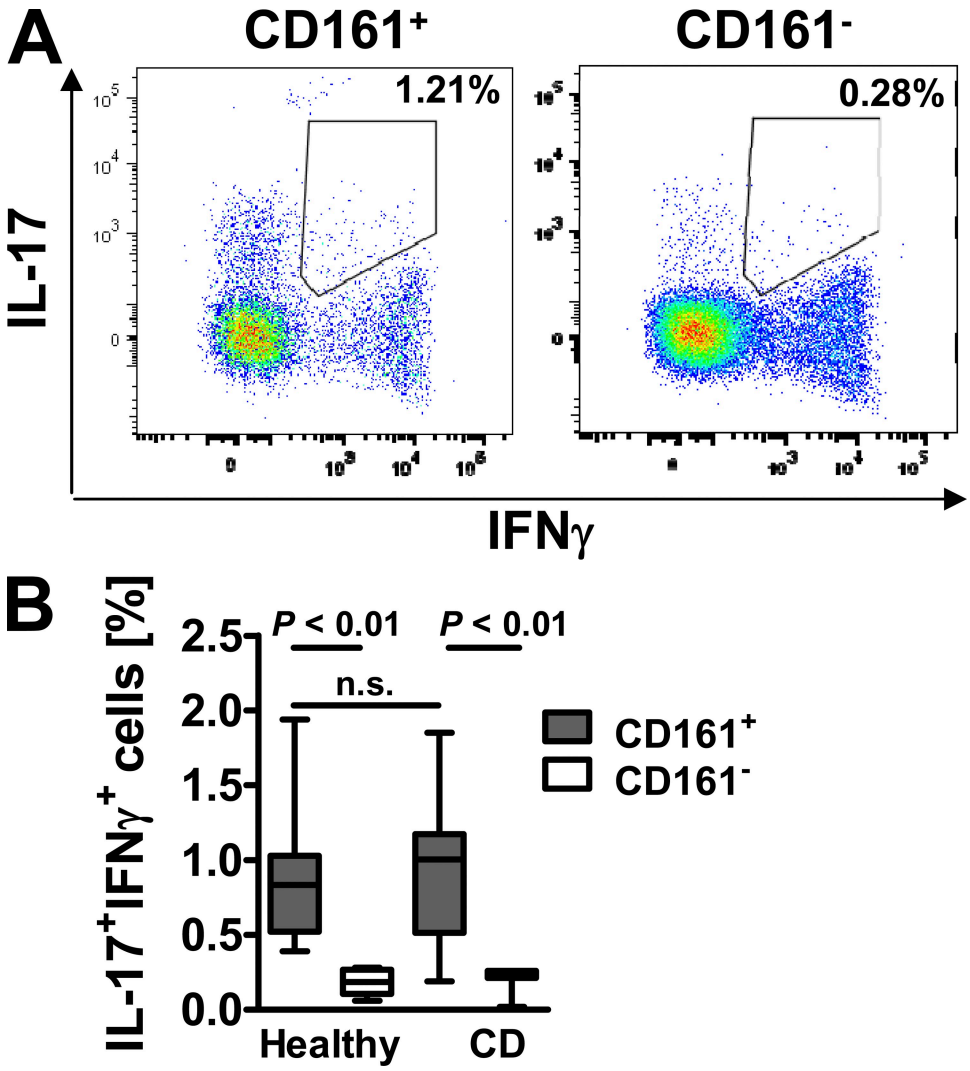
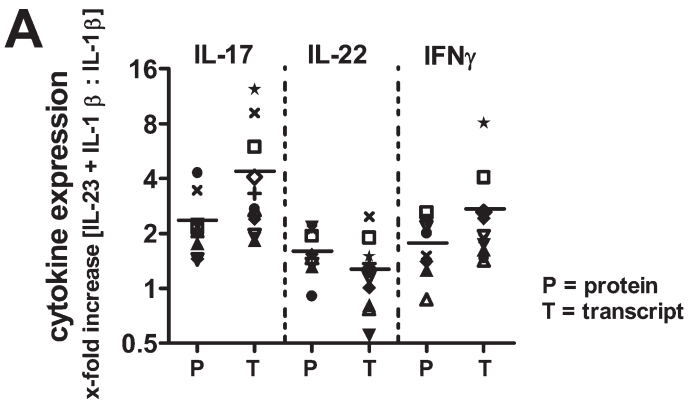


Figure S2. IL-17 and IFN-γ double producers are contained within the CD161⁺ CD4 T memory cell compartment. $CD4^+CD25^-CD45RA^-$ T memory cells were FACS purified from PBMCs from healthy donors and CD patients. Cells were cultured with PMA and ionomycin for 4.5 h, and were stained for CD161 as well as IL-17 and IFN-γ. Representative FACS plots show cytokine production during CD. Box plots presents cytokine-positive cells as a percentage of $CD161^+$ or $CD161^-$ CD4 T memory cells in healthy and CD donors, as indicated. Boxes show interquartile range, the whiskers show minimum to maximum. Eight different donors per disease state are shown.



B

	IL-17		IL-22		IFNγ	
	protein	transcript	protein	transcript	protein	transcript
mean	2.36	4.40	1.60	1.28	1.77	2.72
median	2.10	2.73	1.50	1.25	1.76	1.94

Figure S3. IL-23 augments IL-1β–induced cytokine expression. $CD161^+$ $CD45RA^-CD25^-CD4^+$ T memory cells were FACS purified from normal PBMCs and cultured for 3 d with IL-1β with or without IL-23. Protein and transcriptional levels of IL-17, IL-22, and IFN-γ were assessed (compare with the data in Fig. 3, B and E), and the x-fold increase in cytokine expression was calculated for IL-23–stimulated cultures for each donor (A) and is displayed as the mean and median fold increase (B). Data from 8 (protein) or 11 (transcript) donors from independent experiments are shown. Horizontal bars in A represent medians.

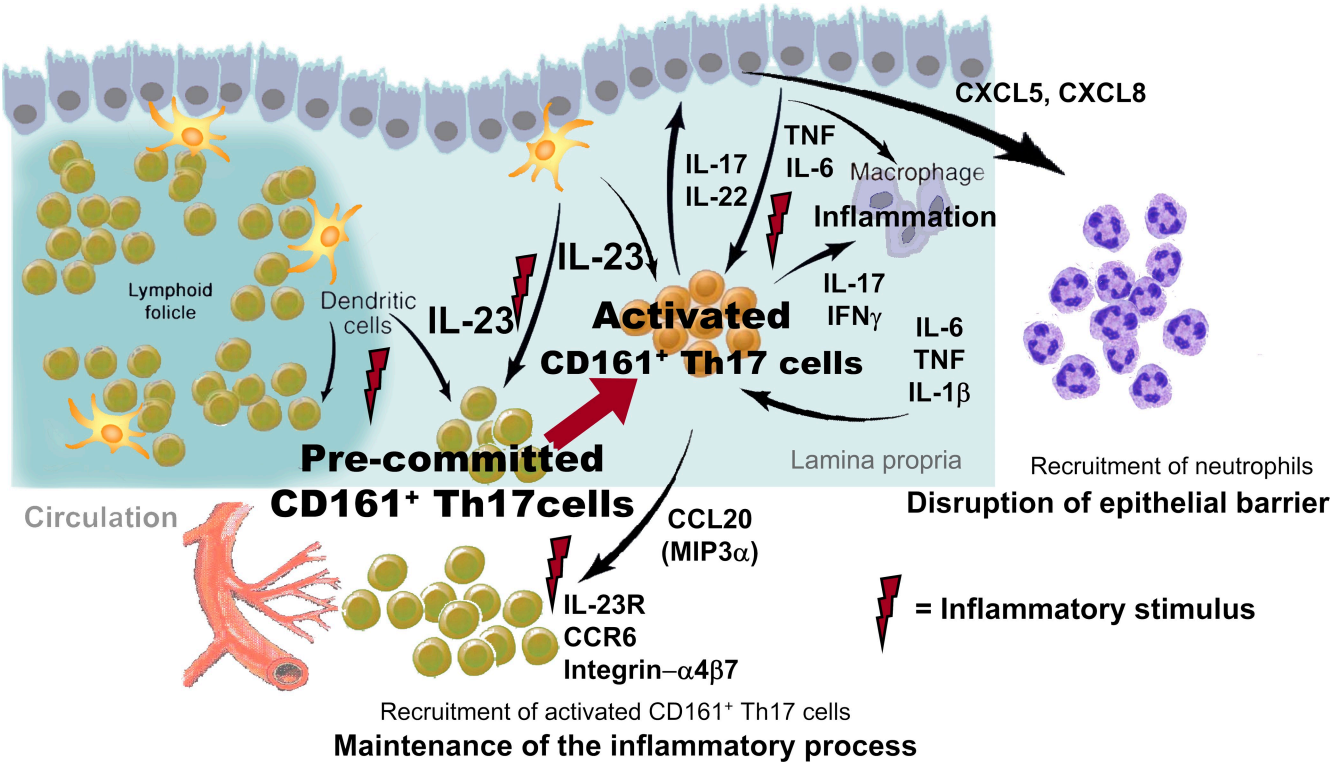


Figure S4. IL-23–inducible CD161⁺ Th17 cells are central to CD inflammation. CD161⁺ CD4 T memory cells comprise a major gut-resident Th cell subset. Activation in situ by IL-23 in concert with antigen-induced inflammatory stimuli leads CD161⁺ T cells to produce IL-17, IL-22, and IFN-γ, factors that activate intestinal epithelial cells and subepithelial myofibroblasts and macrophages to release chemokines and inflammatory mediators. Subsequent recruitment of neutrophils and activated macrophages mediates tissue damage. Recruitment and further activation of integrin α4β7⁺ CD161⁺ Th17 cells sustains and potentiates tissue inflammation.