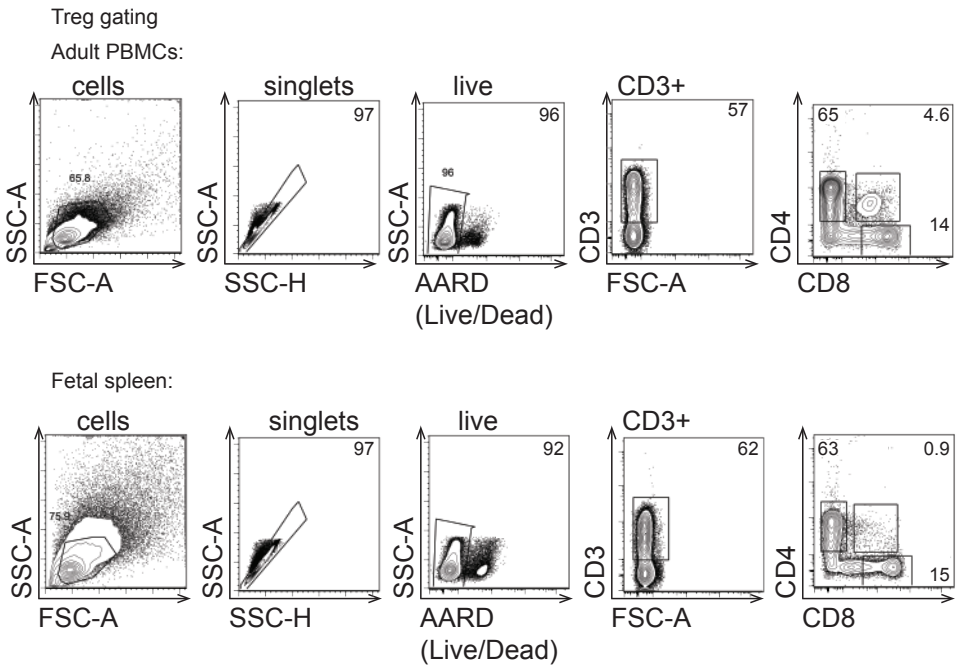
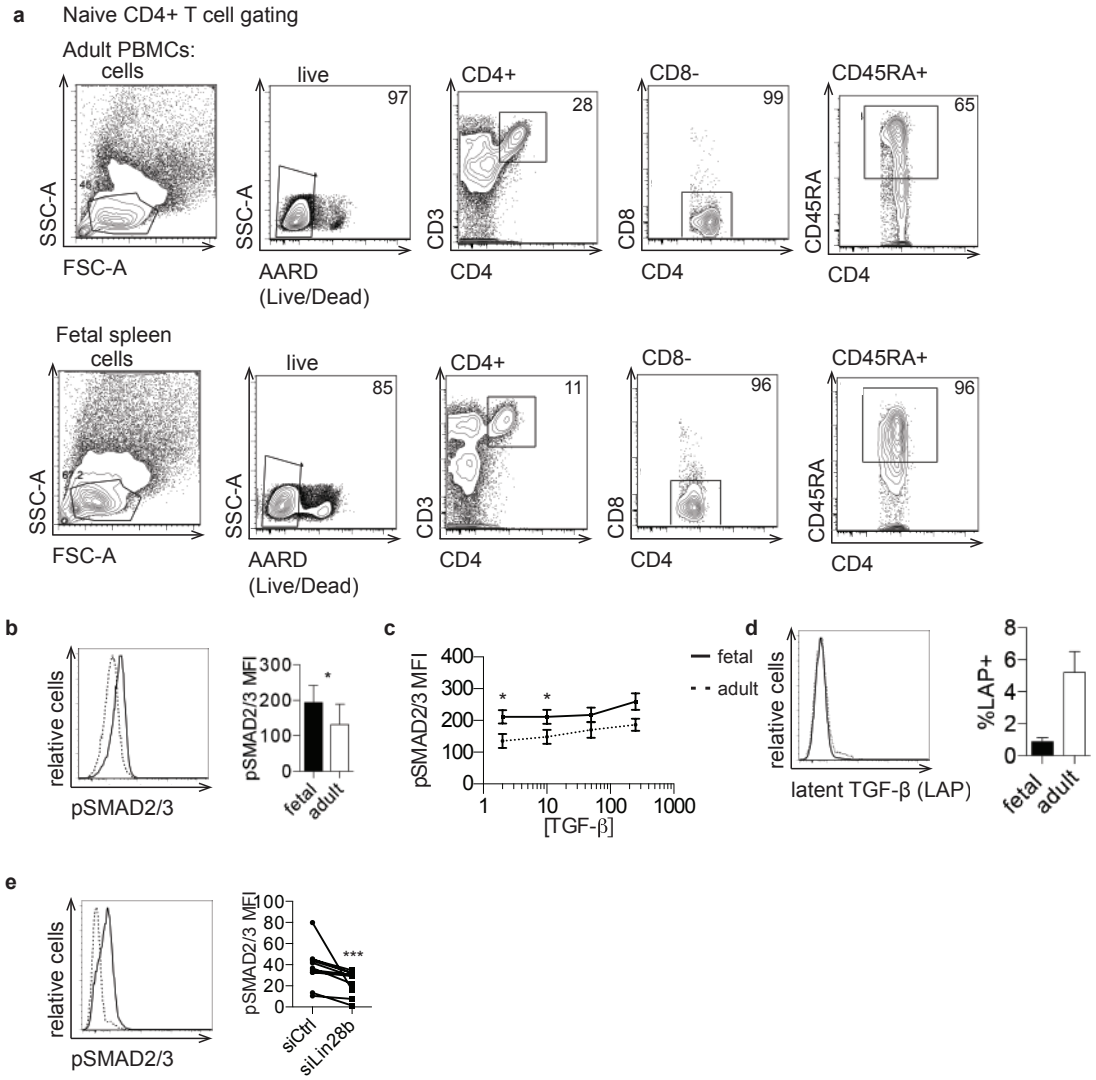
Supplemental Figure 1



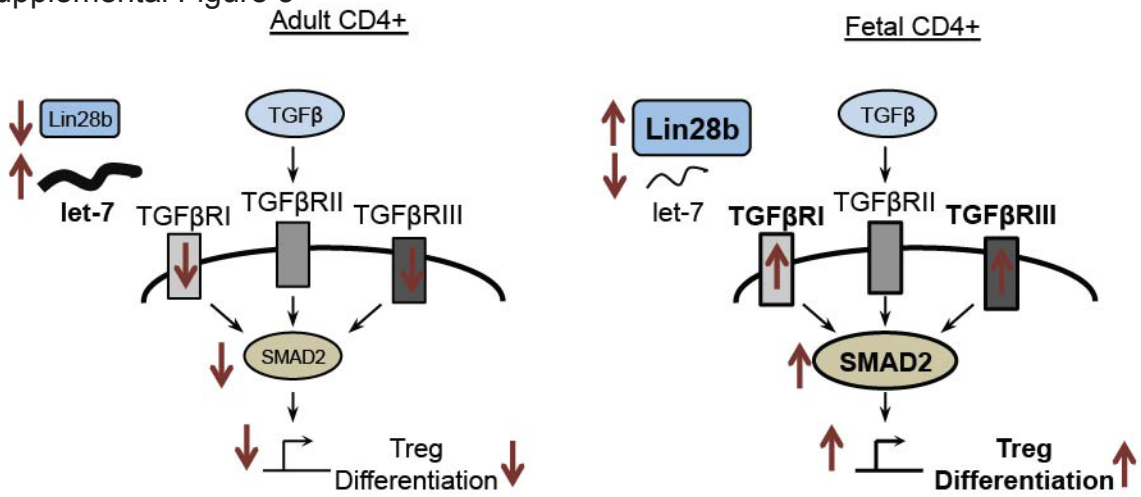
Supplemental Figure 1. Gating strategy for analyzing FoxP3+CD4+ T cells from adult peripheral blood and fetal spleen.

Supplemental Figure 2



Supplemental Figure 2. a. Gating strategy for analyzing naïve CD4+ T cells from adult peripheral blood and feta spleen. b. Flow cytometry analysis of staining for intracellular phosphorylated SMAD2/SMAD3 on fetal and adul FoxP3+CD4+ T cells in the absence of TGF-β. Data are quantified at the right. c. Graph shows phosphorylated SMAD2/3 on fetal and adult naïve CD4+ T cells with increasing TGF-β. d. Flow cytometry analysis of staining fo latent TGF-β (LAP) on fetal and adult T cells. Bar graph show percentage of LAP+ cells within the naïve CD4+ population. e. Flow cytometry analysis of staining for intracellular phosphorylated SMAD2/SMAD3 on siCtrl- and siLin28b-treated CD4+ T cells in the absence of TGF-β.

Supplemental Figure 3



Supplemental Figure 3. Proposed model for Lin28b regulation of fetal Treg differentiation. Lin28b is expressed in human fetal naïve CD4+ T cells. Lin28b expression inhibits let-7 biogenesis, relieving let-7-mediated repression of transcripts including TGF- RI, TGF- RIII, and SMAD2. Increased expression of these proteins promotes more signaling through the TGF- pathway and enhanced Treg differentiation.