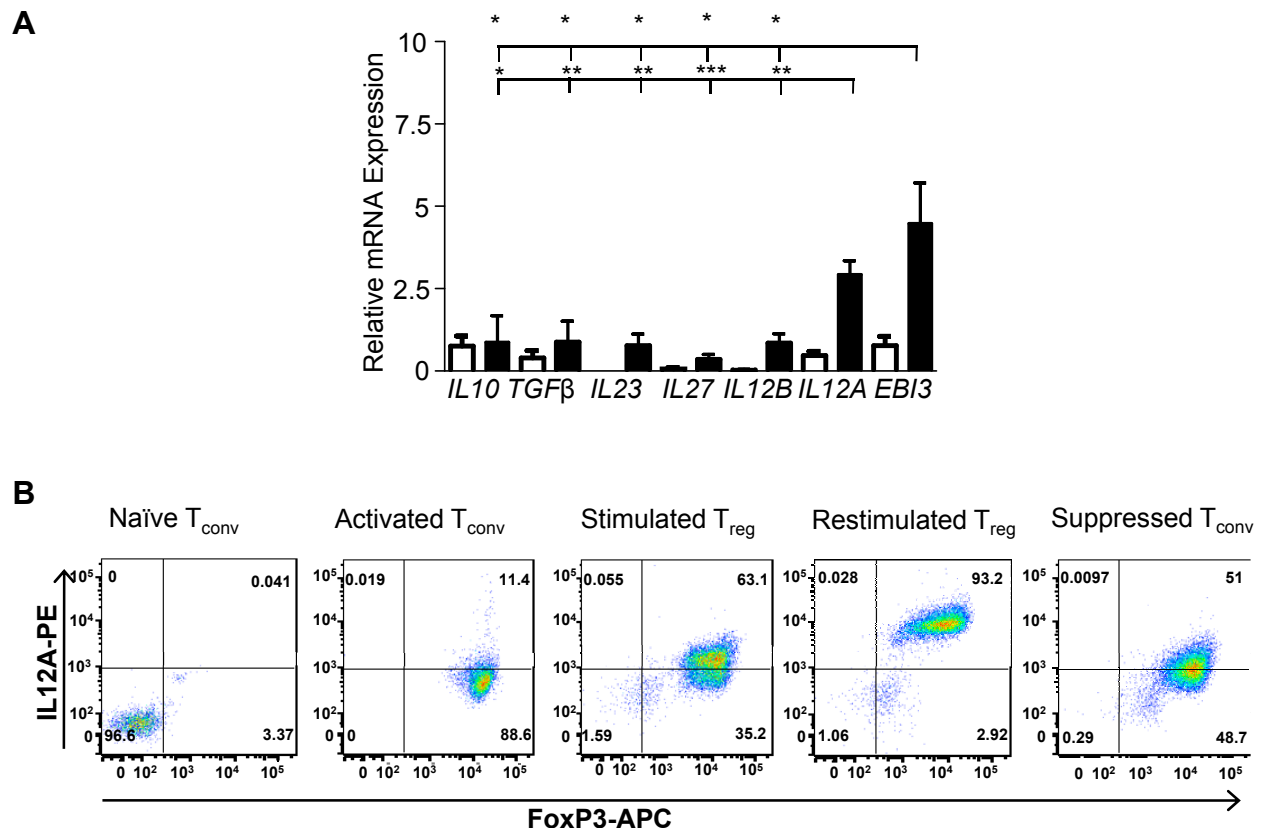
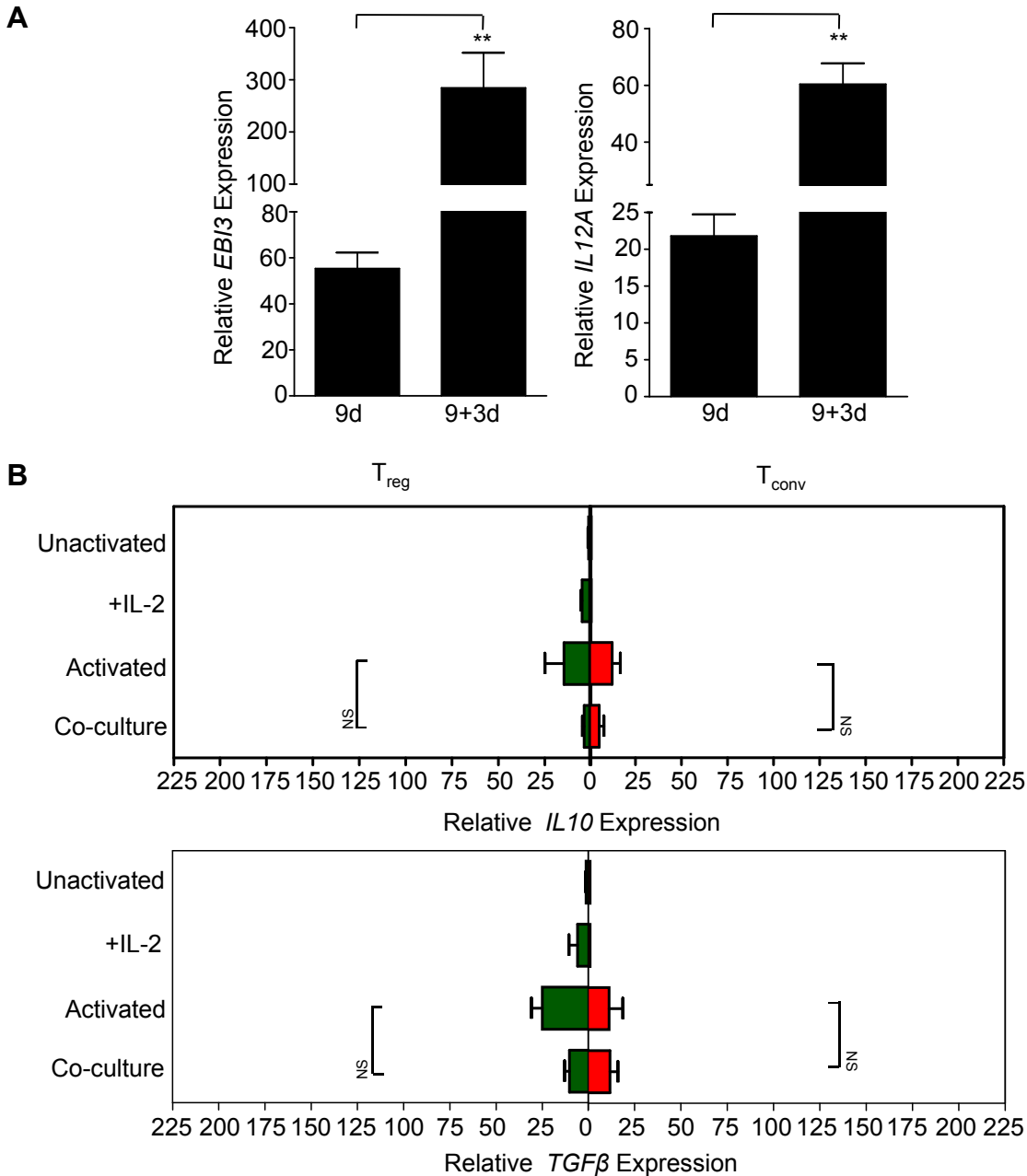


SUPPLEMENTAL TABLE 1: Primers used in this study.

Gene Name	Forward Primer	Reverse Primer
<i>EBI3</i>	5' GCAGCAGACGCCAACGT 3',	5' CCATGGAGAACAGCTGGACAT 3'
<i>IL12A</i>	5' CCTTCACCACTCCCAAAC 3'	5' TGTCTGGCCTTCTGGAGCAT 3'
<i>IL12B</i>	5' TTTTCTGGCATCTCCCCTCGTG 3'	5' GGGTGGGTCAGGTTTGATGATG 3'
<i>TGFB</i>	5' CTGCTGAGGCTCAAGTTAAAAGTG 3'	5' TGAGGTATCGCCAGGAATTGTT 3'
<i>IL27</i>	5' GCGGAATCTCACCTGCCA 3';	5' GGAAACATCAGGGAGCTGCTC 3'
<i>IL23</i>	5' GAGCCTTCTCTGCTCCCTGAT3'	5' AGTTGGCTGAGGCCAGTAG 3'
<i>IL10</i>	5' GCCGTGGAGCAGGTGAAG 3';	5' TGGCTTTGTAGATGCCTTTCTCT 3'



SUPPLEMENTAL FIGURE S1: (A) T_{conv} and T_{reg} cells were isolated from PBMCs based on $CD4^+CD25^+CD45RA^+$ (T_{conv}) [open bars] and $CD4^+CD25^+CD45RA^-$ (T_{reg}) [closed bars] expression and expanded for 9d using anti-CD3/anti-CD28-coated latex beads and IL-2 (500 IU/ml) for T_{regs} and IL-2 (100 IU/ml) for T_{conv} . After 9d, RNA was isolated, cDNA generated and qPCR analysis performed. Relative expression of mRNA encoding IL10, TGF β , IL23, IL27, IL12B, IL12A and EBI3 was determined. Data represent the mean \pm SEM of 6 independent experiments [* $p < 0.05$, ** $p < 0.005$ and *** $p < 0.001$]. Results are presented relative to those of naïve T_{conv} cells. (B) Indicated cell populations were stained for FoxP3 and IL-12A expression. Cells were fixed and permeabilized using 8E buffer and then stained for IL-12A. This was followed by FoxP3 staining (eBioscience, San Diego, CA). Representative dot plots are shown. Naïve and Activated T_{conv} were used as control. Foxp3 and IL-12 A expression in $CD4^+$ gated cells. Stimulated T_{reg} refer to 9 day activated T_{regs} . Restimulated T_{reg} refer to 9+3 day activated T_{regs} . Suppressed T_{conv} are the T_{conv} isolated for cocultures.



SUPPLEMENTAL FIGURE S2: (A) Restimulation of T_{reg} cells after 9d leads to increased *EB13* and *IL12A* expression. Human T_{conv} and T_{reg} cells were purified from umbilical cord blood by FACS based on cell surface expression of CD4 and CD25. Purified cells were expanded for 9 d, using anti-CD3/anti-CD28-coated latex beads and IL-2 (500 IU/ml). The cells were restimulated for additional 3 d, with anti-CD3/anti-CD28-coated latex beads and IL-2 (100 IU/ml) after the initial 9 d stimulation. RNA was isolated, cDNA generated and qPCR analysis performed. Relative *EB13* and *IL12A* expression. Data represents the mean \pm SEM of 5 independent experiments [** $p < 0.005$]. Results are presented relative to those of T_{conv} cells. The T_{conv} cells were expanded similarly to T_{reg} cells except that IL-2 was used at a final concentration of 100 IU/ml for the initial 9 d expansion. (B) Co-culturing T_{reg} and T_{conv} cells does not lead to substantial up-regulation of either IL-10 or TGF β in either population. Activated T_{reg} cells (prepared as described in Fig. 1) were labeled with eFluor[®]670 and cultured with CFSE-labeled naïve T_{conv} cells in the presence or absence of IL-2, anti-CD3/CD28-coated latex beads. At the end of 3 d, cells were purified by FACS. RNA was extracted and cDNA was generated from the indicated populations. Relative *IL10* (upper panel) and *TGF β* expression (lower panel) was determined by qPCR. Data represents the mean \pm SEM of 5 independent experiments [* $p < 0.05$, ** $p < 0.005$, and *** $p < 0.001$]. Results are presented relative to those of naïve T_{conv} cells.