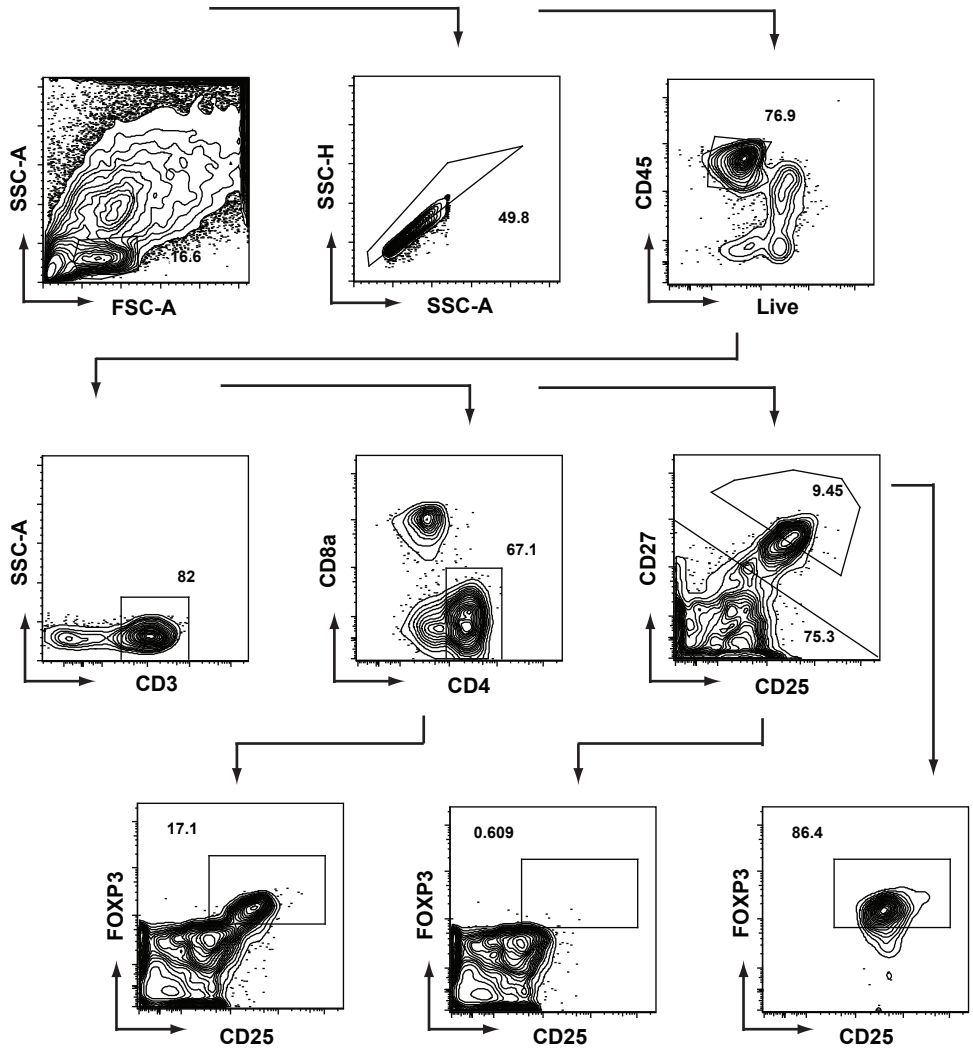
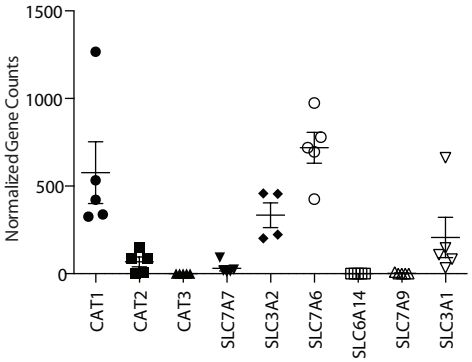
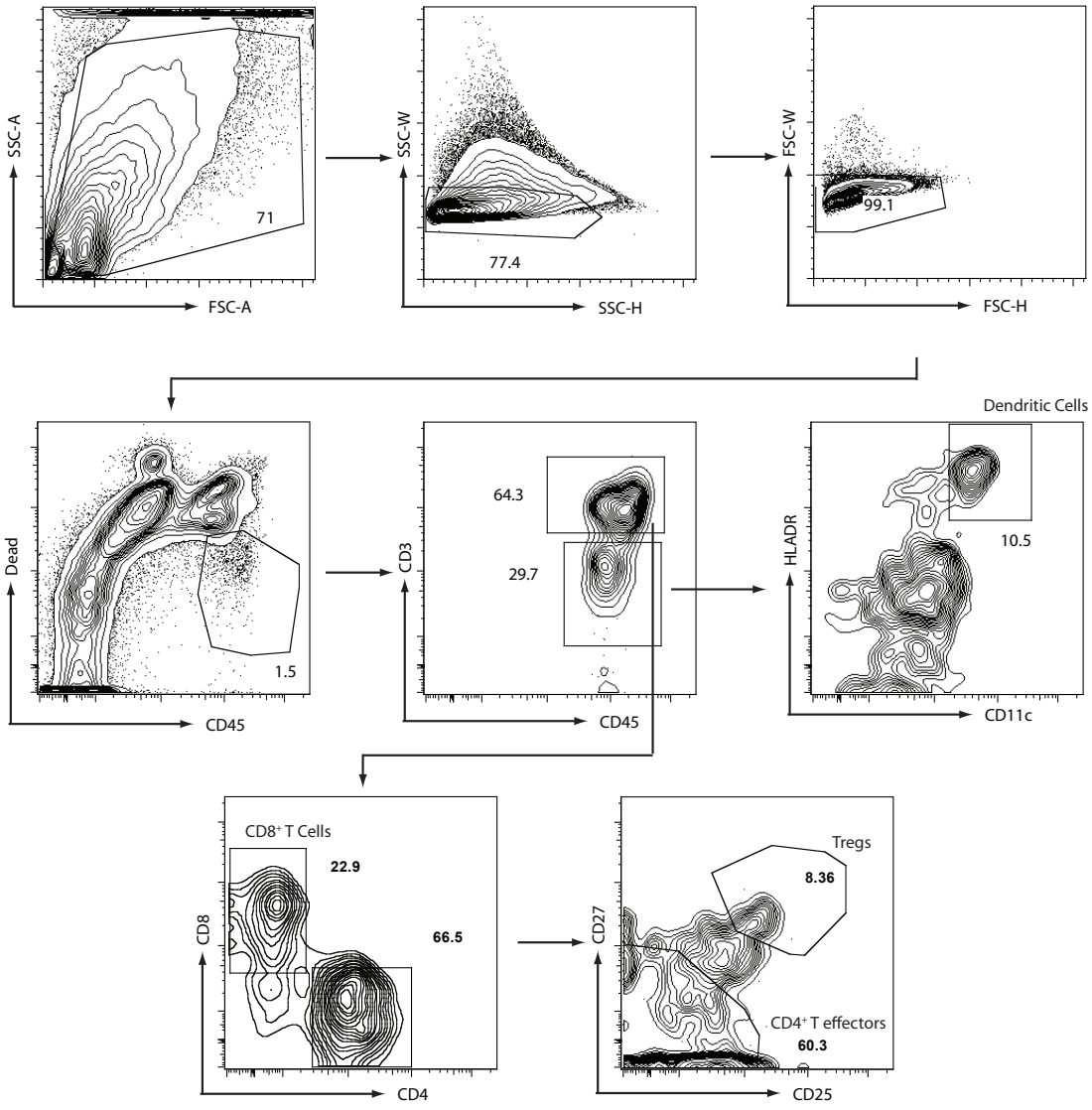
Supplemental Figures



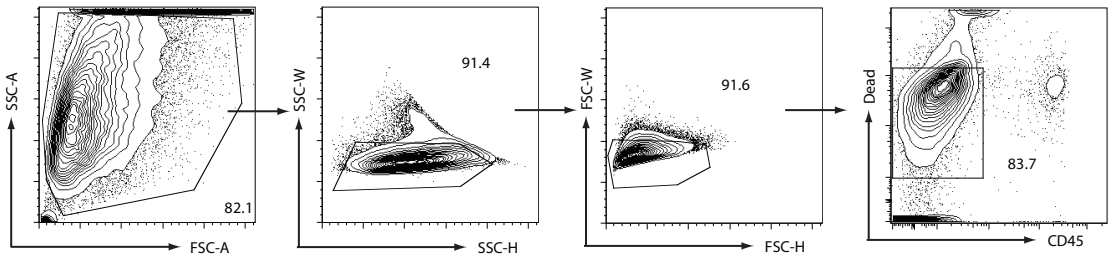
Supplementary Figure 1: Gating strategy of purification of skin Tregs ($CD45^+CD3^+CD4^+CD8^-CD25^+CD27^+$) and Teffs ($CD45^+CD3^+CD4^+CD8^-CD25^-CD27^-$) based upon FOXP3 staining of a separately stained control.



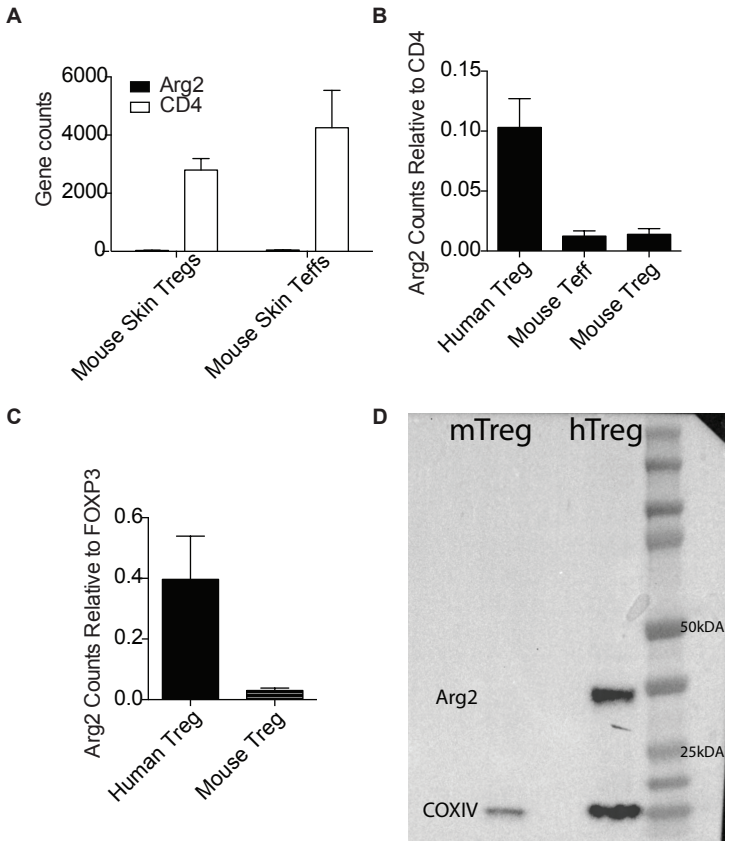
Supplementary Figure 2: Normalized counts of arginine transport system genes in Tregs (n=5) purified from healthy human skin. Error bars are mean ± s.e.m.



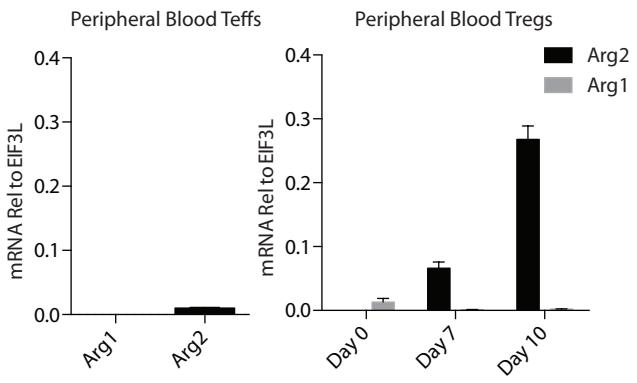
Supplementary Figure 3: Flow cytometric gating strategy for purification of dendritic cells ($CD45^+CD3^-CD11c^+HLADR^+$), $CD8^+$ T cells ($CD45^+CD3^+CD4^-CD8^+$), Tregs ($CD45^+CD3^+CD4^+CD8^-CD27^{hi}CD25^{hi}$) and $CD4^+$ Teffs ($CD45^+CD3^+CD4^+CD8^-CD27^{low}CD25^{low}$) from normal human skin.



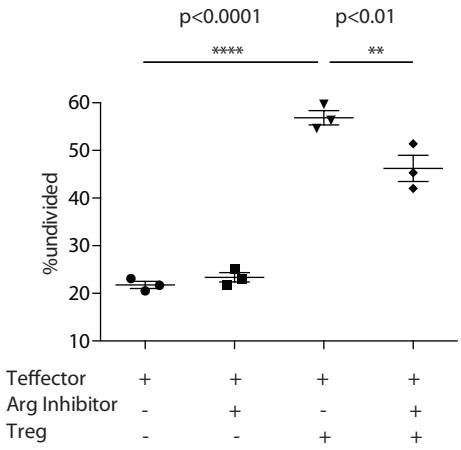
Supplementary Figure 4: Flow cytometric gating strategy for purification of skin keratinocytes (live, singlet, $CD45^-$) isolated from epidermal cell suspension from normal human skin.



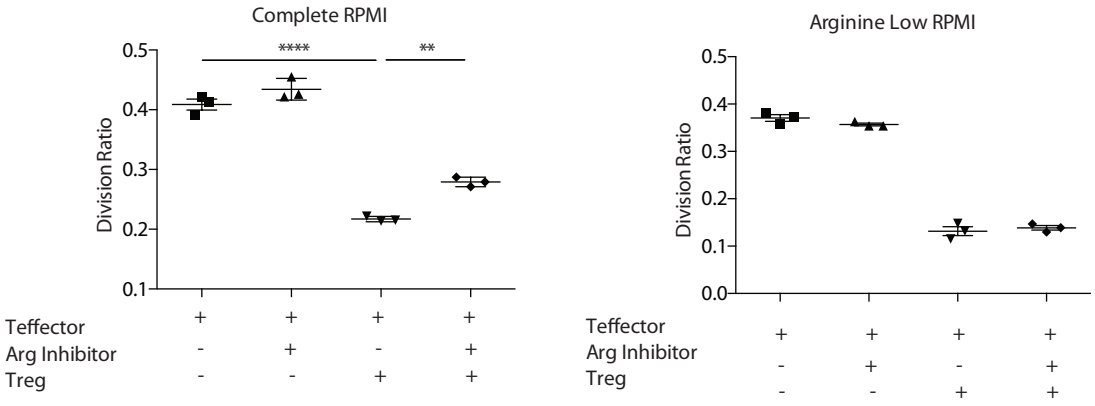
Supplementary Figure 5: A) Arg2 raw counts from RNAseq data from mouse Tregs (n=4) and $CD4^+$ Teffectors (n=2) sort-purified from mouse skin. **B)** Arg2 counts normalized to counts of CD4, comparing data from human skin Tregs to mouse skin Tregs and skin $CD4^+$ Teffectors. **C)** Arg2 counts normalized to counts of FOXP3, comparing data from human skin Tregs to mouse skin Tregs. Error bars are mean ± s.e.m. **D)** Human Tregs and mouse Tregs were expanded for fourteen days *in vitro*. 500,000 cells from each condition were lysed, run on an SDS gel, and blotted with anti-Arg2 and anti-COXIV as a housekeeping gene.



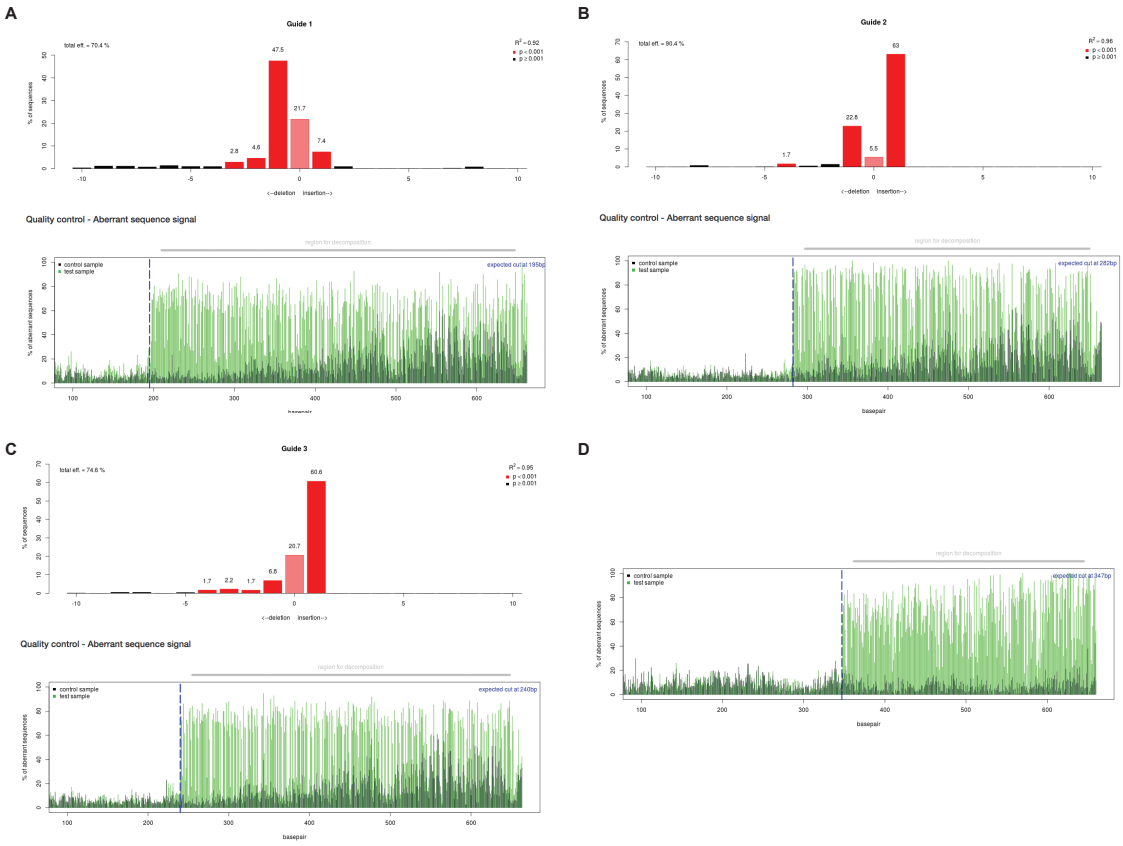
Supplementary Figure 6: qRT-PCR for Arginase 1 and Arg2 mRNA expression in unstimulated sort-purified peripheral blood $CD4^+$ Teff cells and in sort-purified peripheral blood Tregs that were unstimulated (Day 0) or stimulated with anti-CD3/anti-CD28 coated beads for 7 or 10 days.



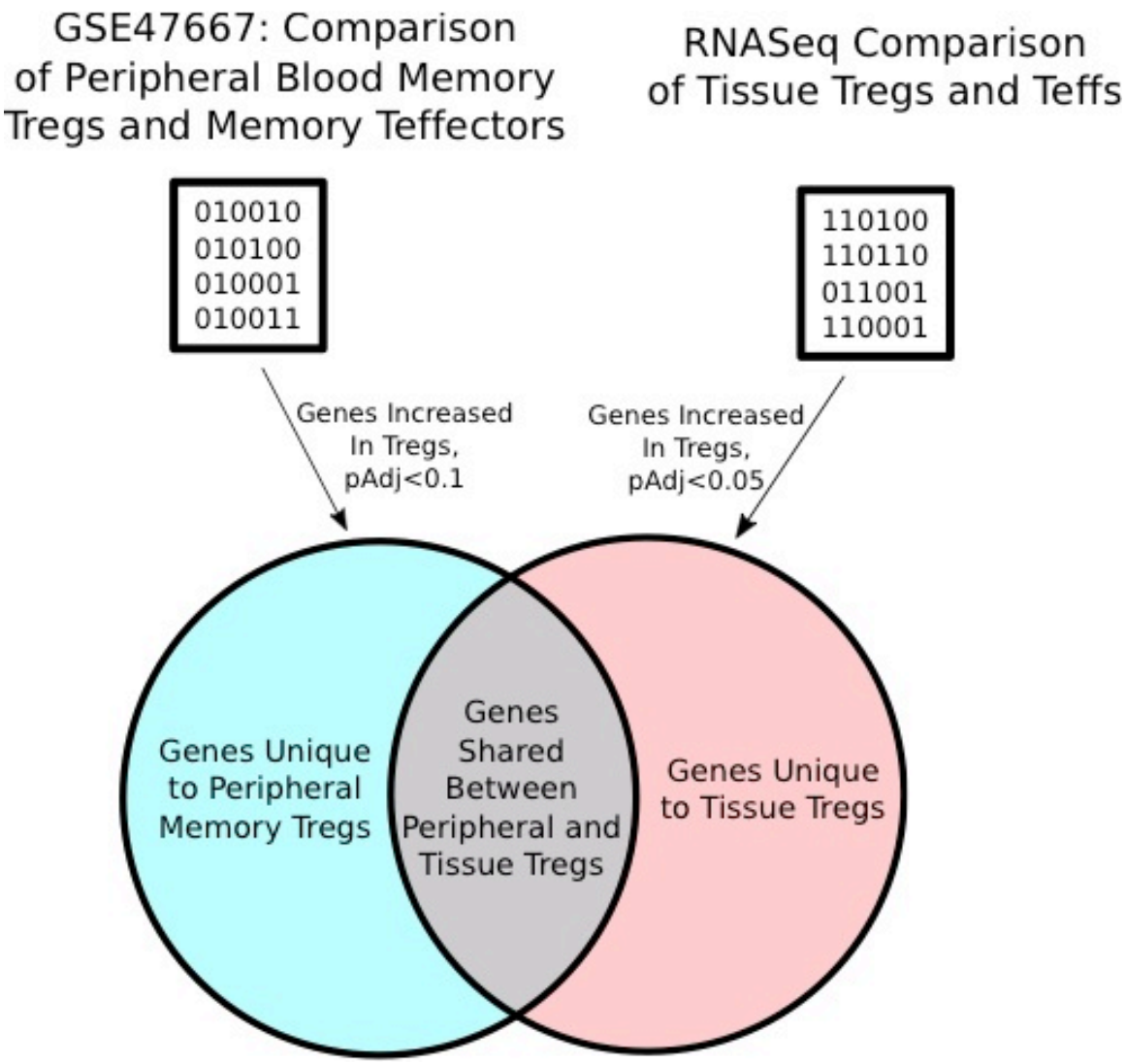
Supplementary Figure 7: Reversal of Treg-mediated suppression of Teff proliferation by addition of the arginase inhibitor, BEC (S-(2-boronoethyl)-L-cysteine) (25 μM), (**** p<0.0001, ** p<0.01, Ordinary 1 Way ANOVA). Experiment was performed with three technical replicates and is representative of three independent experiments. Error bars are mean ± s.e.m.



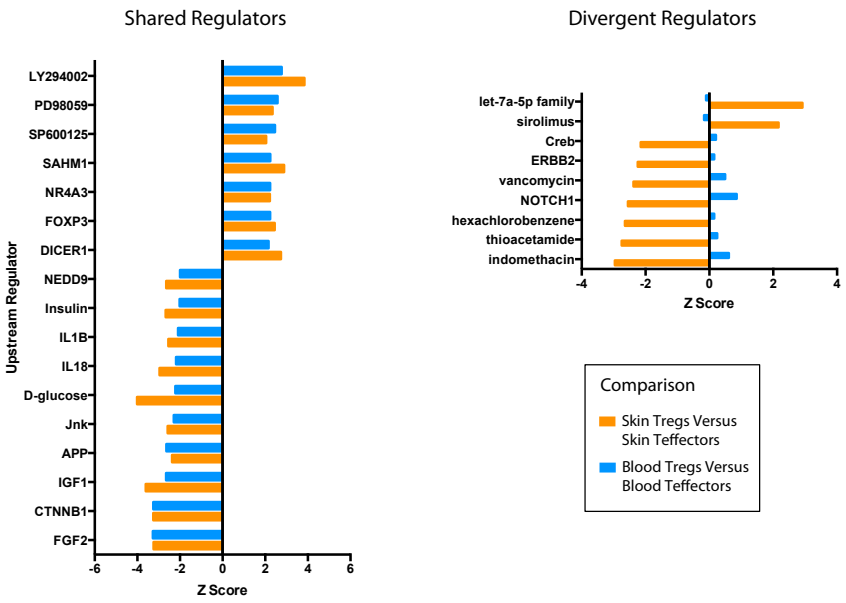
Supplementary Figure 8: Reversal of Treg-mediated suppression of Teff proliferation by addition of an arginase inhibitor (BEC, S-(2-boronoethyl)-L-cysteine, 100 μM) (left) is attenuated in arginine-low media (right), (**** $p<0.0001$, ** $p<0.01$, Ordinary 1 Way ANOVA). Experiment was performed with three technical replicates and was performed once. Error bars are mean ± s.e.m.



Supplementary Figure 9: TIDE (Tracking Indels by Decomposition) Analysis of DNA editing following CRISPR mediated Arg2 deletion for guides 1 (A), 2 (B), 3 (C), and pooled (D). DNA sequences from bulk cells treated with Arg2 guide RNAs (green) are compared to those treated with scrambled guide RNA control (black). The blue line shows the expected cut site for the lead guide RNA; following the expected cut site an increase in the frequency of aberrant nucleotides in the Sanger sequencing is seen for Arg2-deleted cells.



Supplementary Figure 10: Generation of a Healthy Human Tissue Treg gene set.



Supplementary Figure 11: Original upstream regulator nomenclature from IPA.