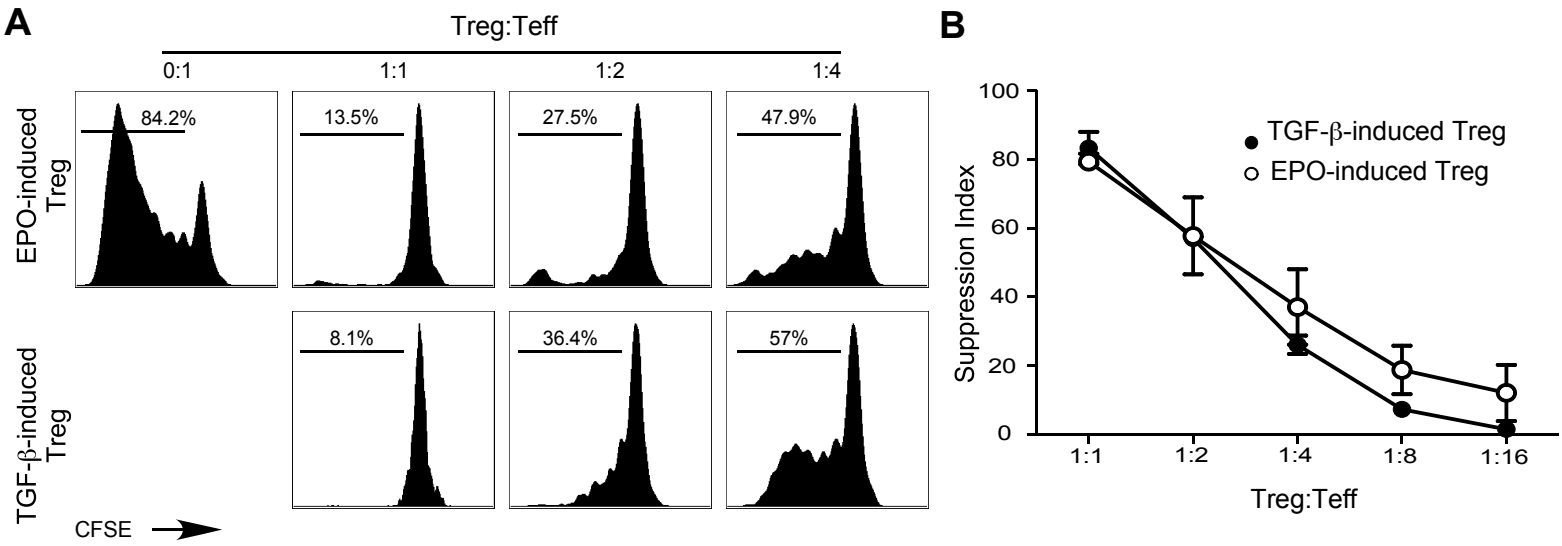
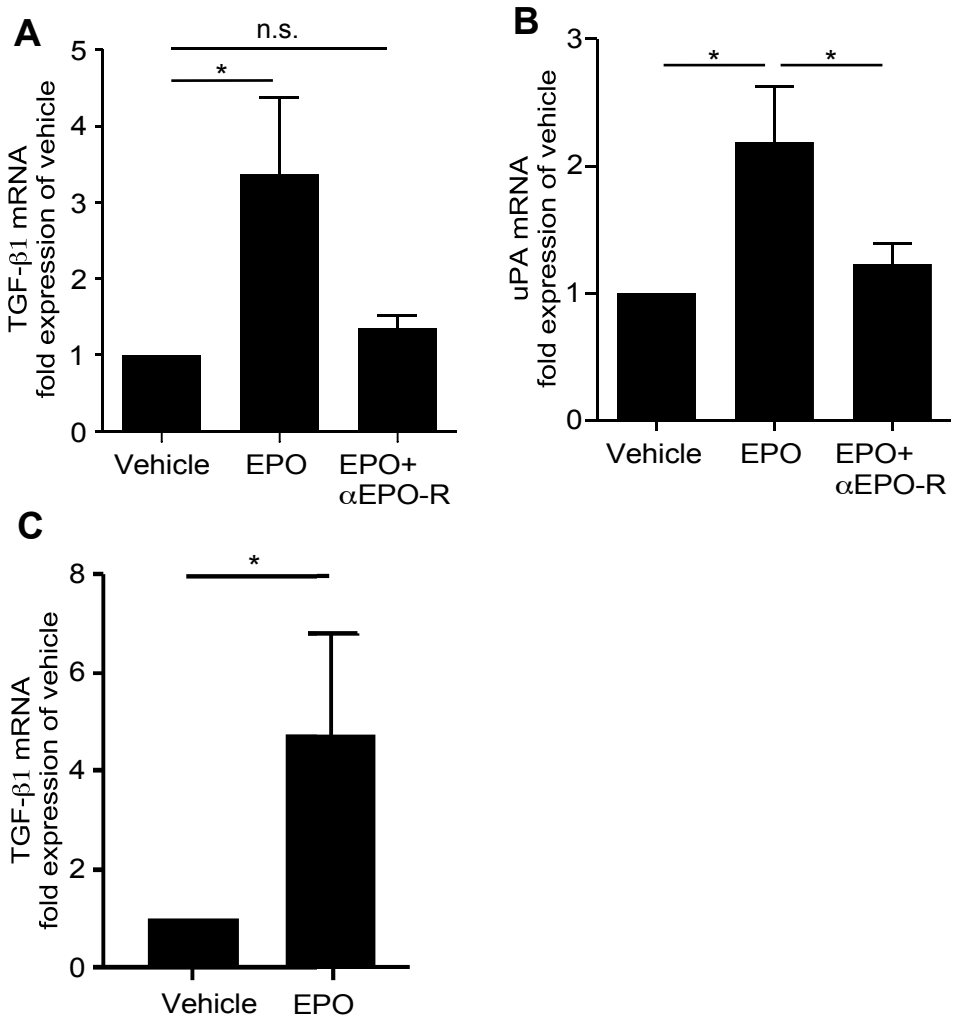
Patients characteristics at study enrolment.

	Patients (n=17)	Controls (n=15)
Age (yrs)	67±12	58±13
Male gender (n; %)	12 (71%)	13 (86%)
SBP (mmHg)	143±20	139±17
DBP (mmHg)	78±16	86±12
CKD Ethiologies		
HTN/DM	62%	40%
GN	20%	40%
Others	18%	18%
eGFR (ml/min/1.73m^2)	15±6	17±6
Hb (g/dl)	9.7±2.8	13.5±1.6
Proteinuria (mg/24h)	70	30

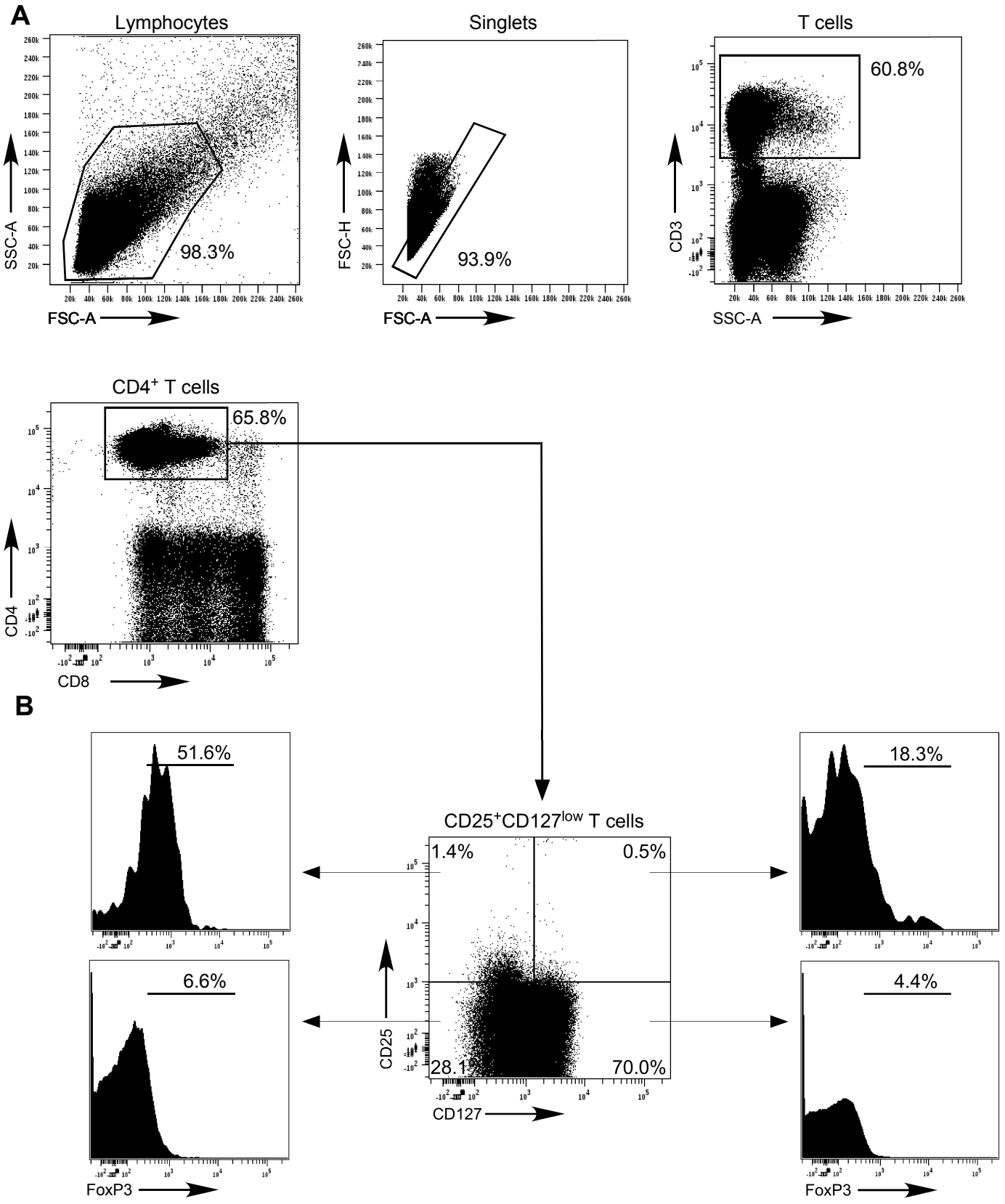
SBP: systolic blood pressure; DBP: diastolic blood pressure; CKD: chronic kidney disease; HTN: hypertension; DM: diabetes mellitus; GN: glomerulonephritis; eGFR: estimated glomerular filtration rate; Hb: hemoglobin.



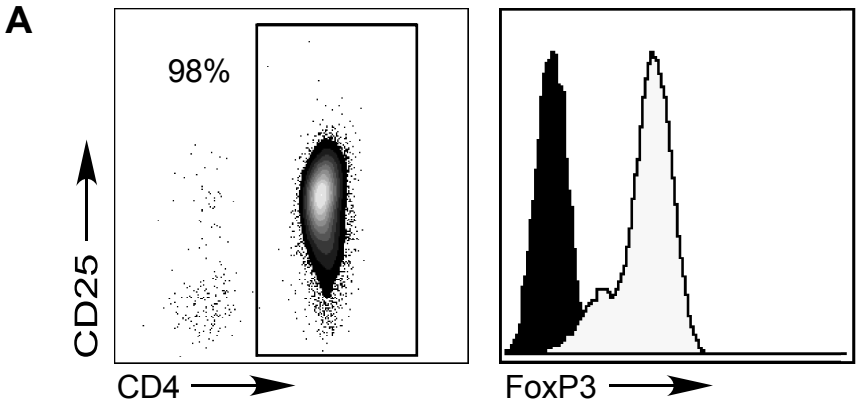
Supplemental Figure 1. Treg induced in the presence of EPO are functional. A) Treg induced in the presence of monocytes, anti-CD3, IL-2 and EPO were enriched by magnetic bead isolation on the basis of CD25 expression (purity >95%) and tested in suppression assays with anti-CD3 stimulated CFSE-labeled responder PBMC (top row) at Teff/Treg ratios indicated. Enriched iTreg resulting from naïve $CD4^+$ T cells cultured with anti-CD3/anti-CD28 coated beads, IL-2 and TGF-β were used as positive controls (bottom row). Representative flow plots are shown gated on responding $CD8^+$ T cells. **B**) summarized quantification of CFSE dilution in PBMC (gated on responding CD8+ cells) at the end of the 5-day suppression assays (n=4, mean and s.e.m., no significant differences were observed at any Treg:Teff ratio).

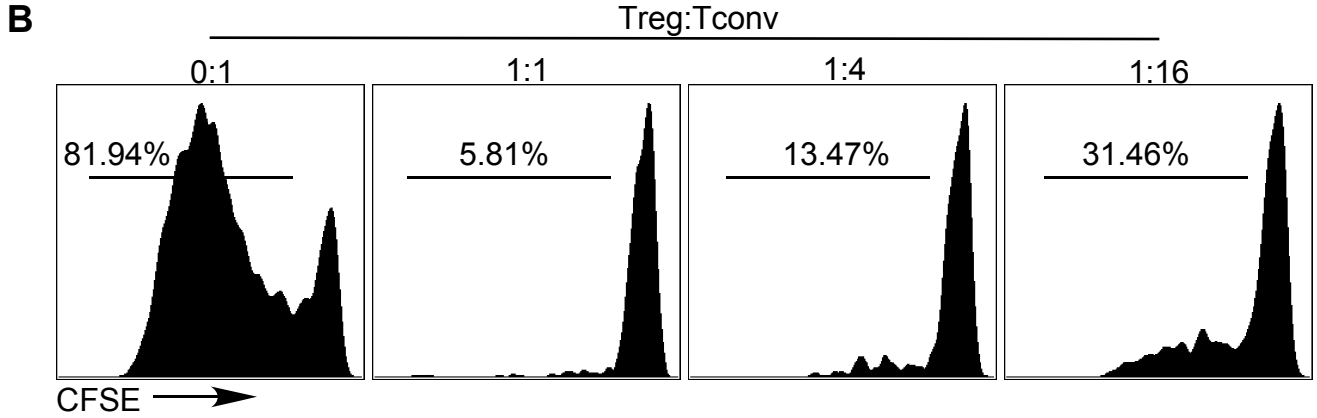


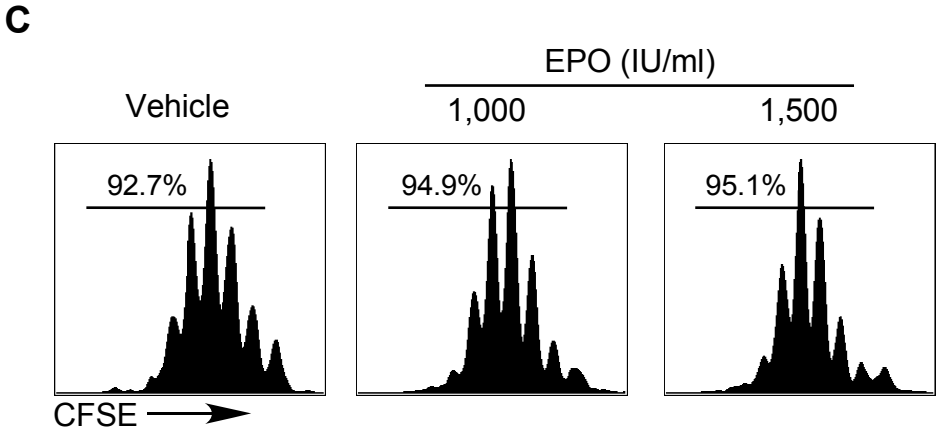
Supplemental Figure 2. EPO promotes TGF-β1 and uPA genes production in human monocytes and tubular cells. A-B) Monocytes were cultured in serum-free media with EPO (1,000 IU/ml) or vehicle control for 2 days. Thereafter, **A**) TGF-β1 and **B**) urokinase plasminogen activator (uPA) genes (qPCR) were determined. **C**) TGF-β1 gene expression in human kidney tubular cells after 2-day culture with EPO or vehicle control. *P<0.05.

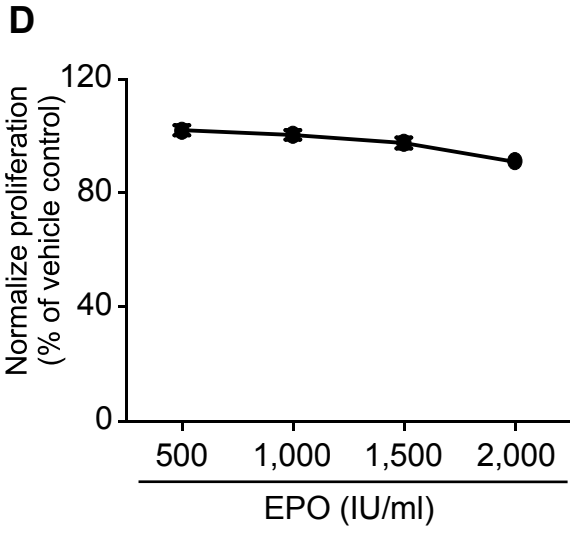


Supplemental Figure 3. Human $CD4^+CD25^+CD127^{low}$ cells are $FoxP3^+$. A) Gating strategy for $CD4^+CD25^+CD127^{low}$ cells; **B)** FoxP3 expression in CD4+ T cells according to the expression of CD25 and CD127. $CD25^+CD127^{low}$ are the ones with the highest expression on FoxP3.

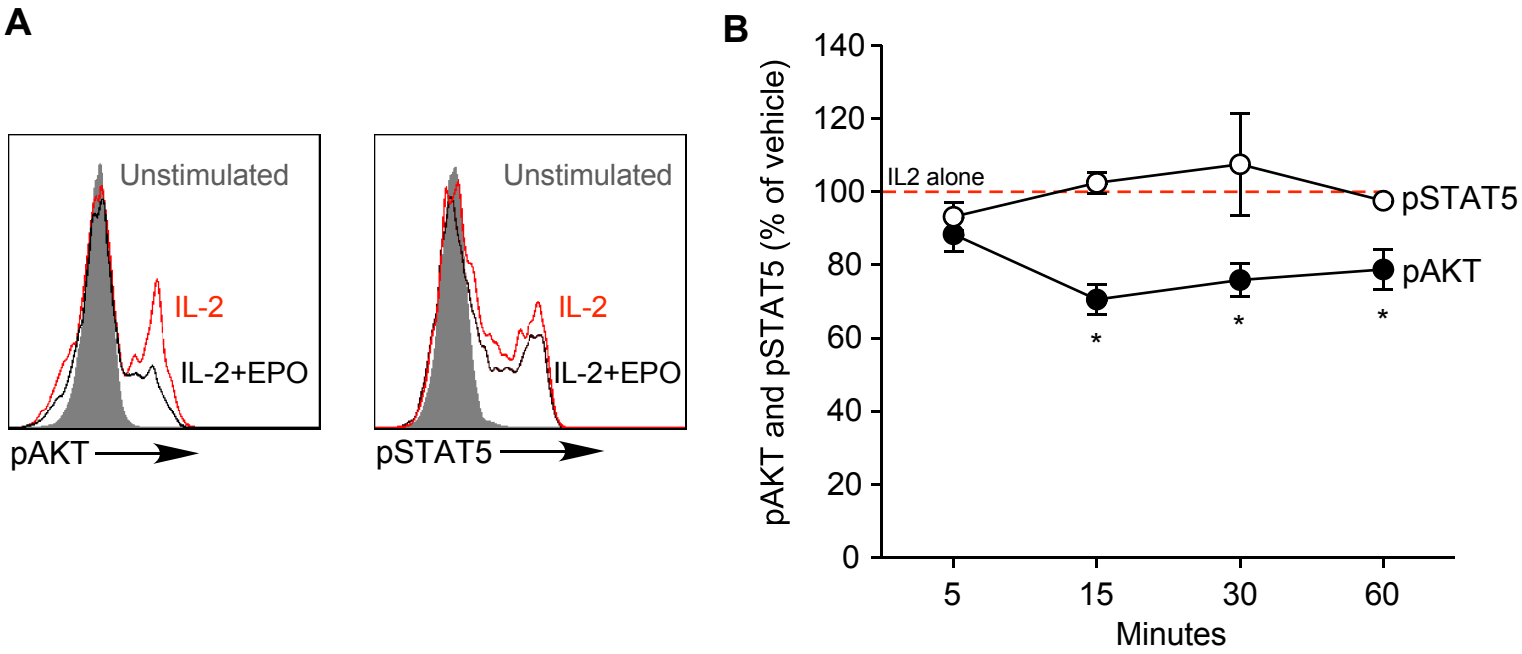




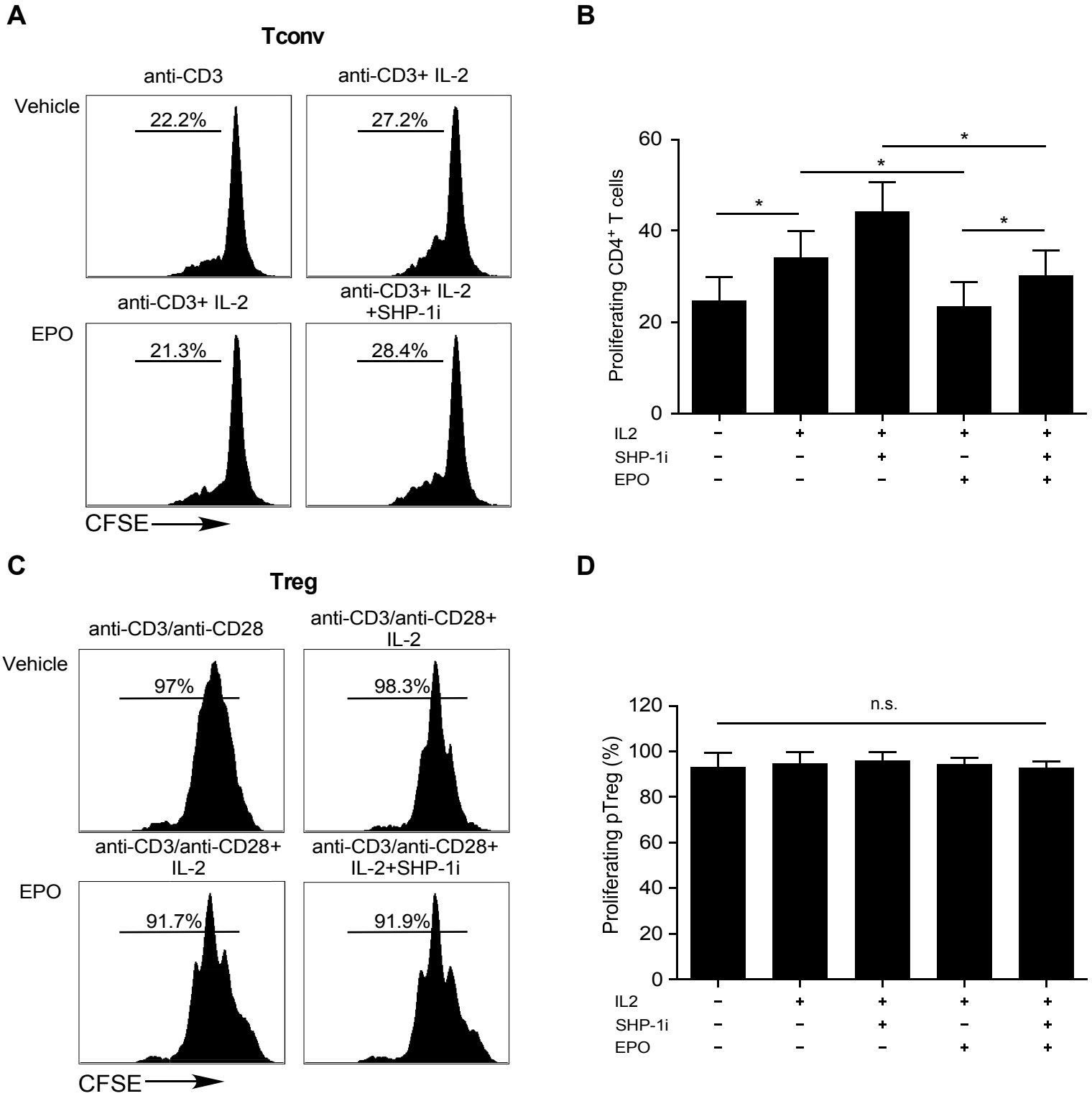




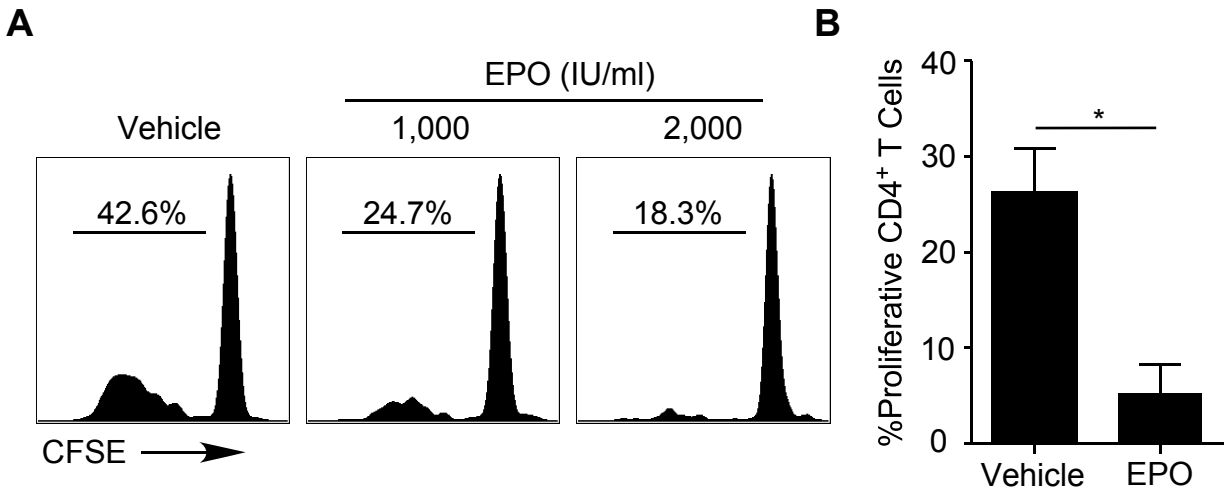
Supplemental Fig. 4. Human pTreg phenotype and function. A) Expanded pTreg were stained for CD4, CD25 and FoxP3 expression. **B**) Treg function was tested in suppression assays with anti-CD3 stimulated CFSE-labeled responder PBMC. A representative suppression assay is depicted. **C-D**) induced Treg were CFSE labeled and activated with anti-CD3, anti-CD28, IL-2 +/- EPO. Representative plots **C**) and data quantification **D**) of CFSE dilution at 5 days.



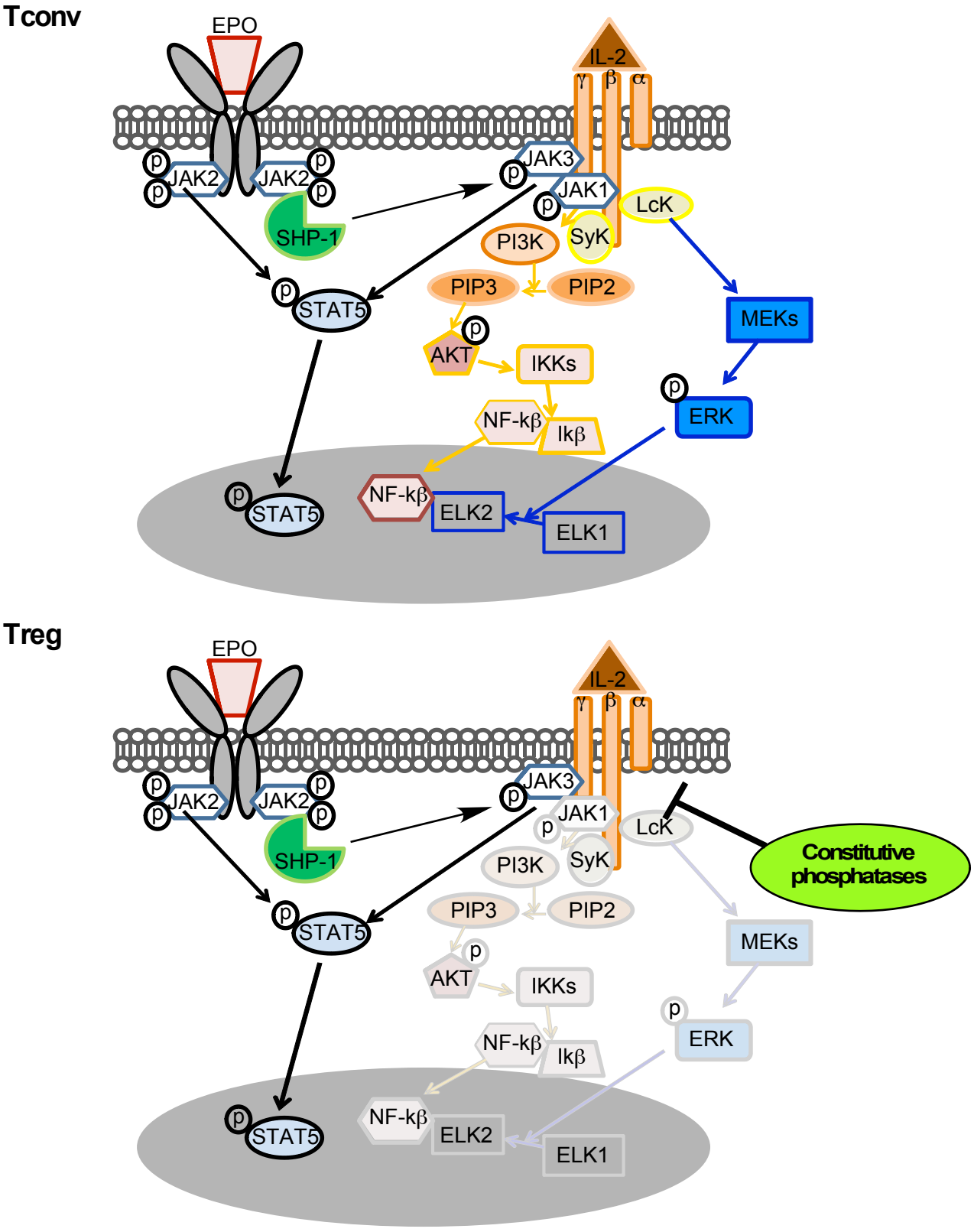
Supplemental Figure 5. EPO uncouples signaling downstream IL-2Rβ but not IL-2Rγ. a-b) Human naive $CD4^+$ T cells were enriched by magnetic bead isolation (purity >95%) and activated with anti-CD3/anti-CD28 coated beads (25 μl/10^6 cells) and IL-2 (100 IU/ml) plus EPO (1,000 IU/ml) or vehicle control for 5-60 minutes. **A**) Representative flow plots and **B**) data quantification show levels of phosphorylated AKT (pAKT) and phosphorylated STAT5 (pSTAT5) assayed by flow cytometry at 30 minutes. The results are expressed as percentage of pAKT or pSTAT5 in EPO treated cells vs. IL-2 alone (set at 100%, red dashed line). Data are from 5-15 independent experiments with different donors (mean and s.e.m.) with >100,000 analyzed events per experiment. *P < 0.05 vs. vehicle at the same time point (One-way ANOVA with Tukey post-test).



Supplemental Figure 6. SHP-1 antagonism rescues EPO induced inhibition of effector T cells. A-B) CFSE-labeled PBMC were activated with anti-CD3 ± IL-2, ± EPO and ± SHP-1 inhibitor (12.5 μg/ml) as indicated for 3 days. **A**) representative flow cytometry plots gated on CD4+ T cells and **B**) summarized quantification of CFSE dilution from 11 independent experiments with different donors. **C-D**) CFSE-labeled $CD4^+$ pTreg were activated for 3 days under the same conditions as in A-B. **C**) Representative flow plots gated on $CD4^+$ T cells and **D**) summarized quantification of 4 independent experiments. *$P < 0.05$ (Student's t-test).



Supplemental Figure 7. Human EPO inhibits murine T cell proliferation. A-B) B6 T cells were enriched by magnetic separation, labeled with CFSE and cultured for 5 days with allogeneic BALB/c APCs plus human EPO (concentrations as indicated) or vehicle control. Representative flow plots gated on CD8+ Tconv **A**) and summarized quantification **B**) of CFSE dilution. Data quantification refers to EPO 2,000 IU/ml (n=6). We obtained similar results when T cells were cultured in the presence of anti-CD3/anti-CD28 (not shown). *P<0.05.



Supplemental Figure 8. Cross-talk between EPOR and IL-2R in T cells. While EPO induces SHP-1 activation in both Tconv and Treg, it only inhibits proliferation in Tconv cells, because it silences signaling downstream IL-2Rβ (important for Tconv, but already silenced by constitutive phosphatases in Tregs), but it leaves signaling downstream IL-2Rg, important for Treg, unaffected.