Supplementary Figures

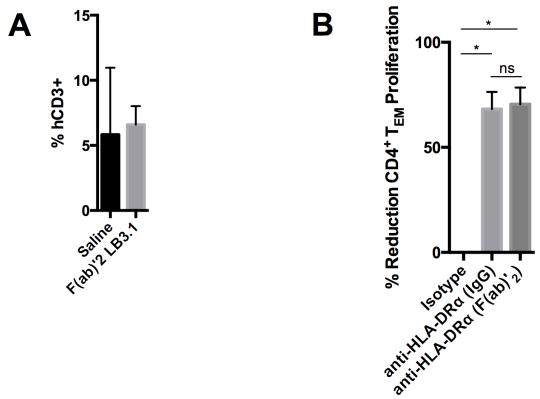

Figure S1: **F(ab)'2 fragments of anti-HLA-DRα mAb does not affect circulating human CD3$^+$ cells *in vivo* but effectively reduces recognition of EC by CD4$^+$ T_{EM} *in vitro***. **(A)** anti-HLA-DRα F(ab)'$_2$ fragment does not affect adoptive transfer of human CD3$^+$ cells to the peripheral circulation of C.B-17 SCID/bg mice (n=3). (**B**) The F(ab)'$_2$ fragments retain blocking activity equivalent to the full length IgG as measured by CD4$^+$ T_{EM} proliferation to allogeneic EC (n=3). Similar results were seen in three independent experiments. * significant by one-way ANOVA with Bonferroni post-hoc test. T_{EM}, effector memory T cell.

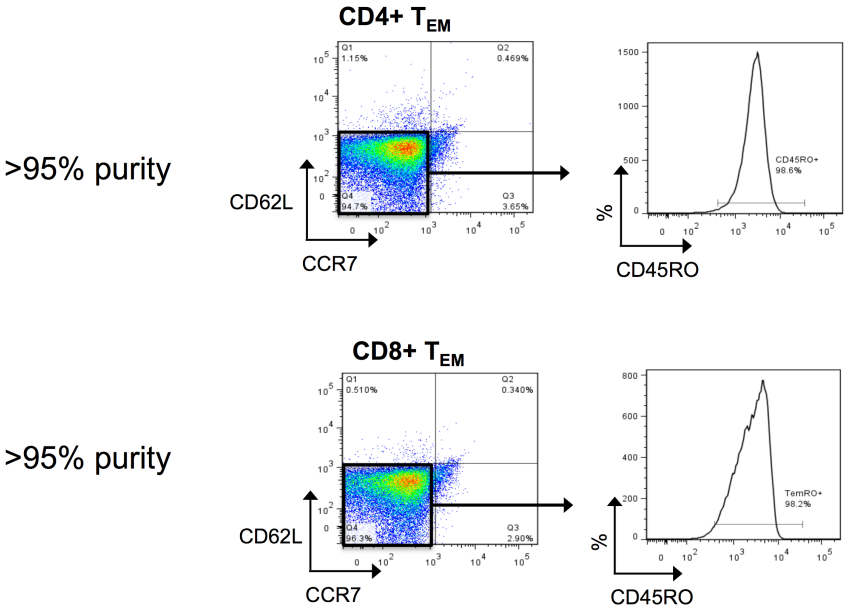

Figure S2: **Magnetic bead immunoselection of $CD4^+$ and $CD8^+$ T_{EM} from human PBMC results in >95% purity for cells that are $CD62L^-CCR7^-CD45RO^+$.** T_{EM}, effector memory T cell.

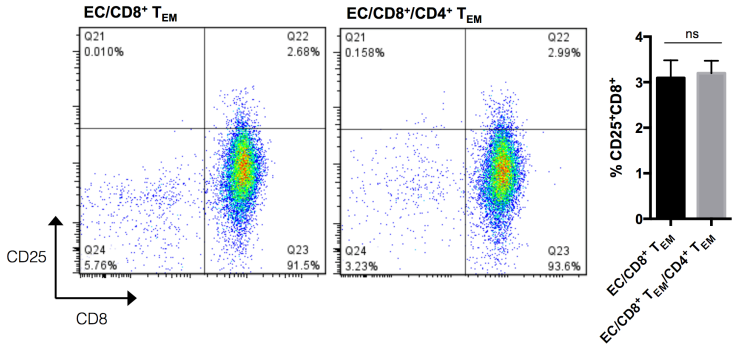

Figure S3: **Addition of CD4$^+$ T_{EM} to EC/CD8$^+$ T_{EM} co-cultures does not significantly increase expression of CD25, a marker of TCR engagement at 48h detected by flow cytometry.** (n=3) Similar results were seen in two independent experiments. * $p<0.05$, 2-tailed Student's t test. T_{EM}, effector memory T cell; TCR, T cell receptor.

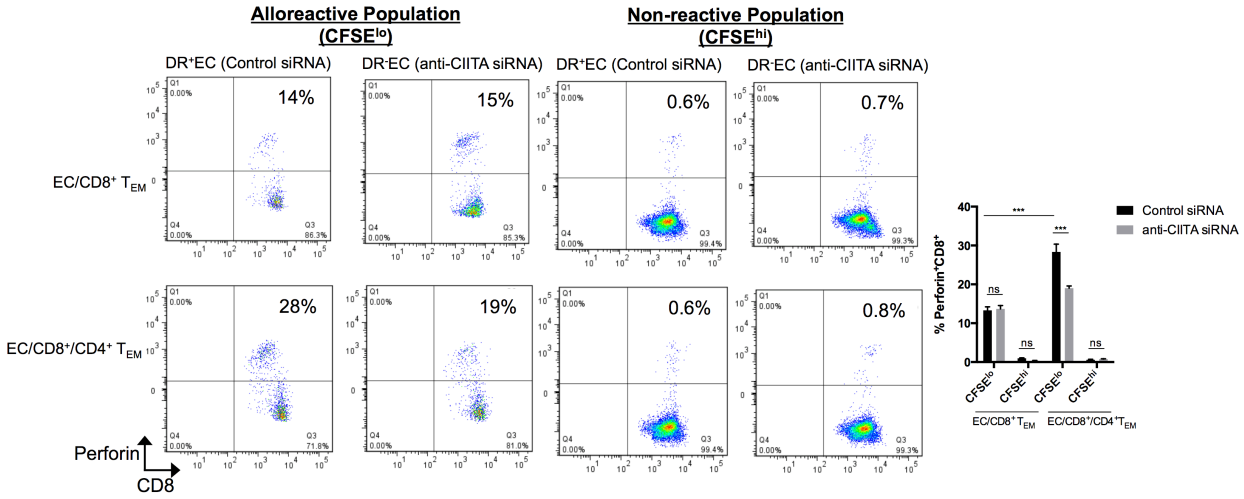

Figure S4: **Concomitant activation of CD4$^+$ T_{EM} increases frequency of perforin$^+$CD8$^+$ T cells in alloreactive population (CFSE^{lo}) but not non-reactive (CFSE^{hi}).** CD8$^+$ T_{EM} alone or CD4$^+$ and CD8$^+$ T_{EM} were cultured with EC that had been pre-treated with anti-CIITA or control siRNA and subsequently activated with IFN-γ, 48h prior. At 7d, cells were harvested and analyzed for CD8$^+$ expression, CFSE dilution and intracellular perforin expression by flow cytometry (n=4). Similar results were seen in three independent experiments. * significant by one-way ANOVA with Bonferroni post-hoc test. T_{EM}, effector memory T cell; CIITA, class II MHC Transactivator.

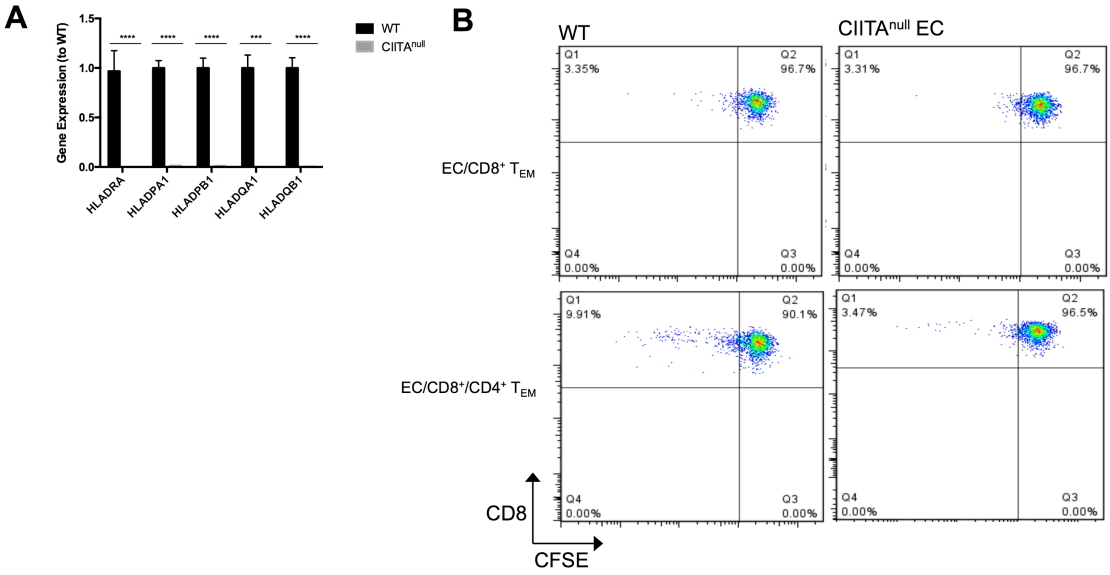

Figure S5: **Genetic ablation of *CIITA* in HECFC-derived EC by CRISPR/Cas9 prevents CD4$^+$ T_{EM} from enhancing CD8$^+$ T_{EM} alloresponses. (A)** Genetic ablation of *CIITA* in EC eliminates expression of class II MHC molecules as measured by qRT-PCR. **(B)** CD8$^+$ T_{EM} or CD8$^+$/CD4$^+$ T_{EM} were co-cultured with unmodified or CIITA^{null} EC pre-treated with IFN-γ to re-induce *CIITA* and measured by CFSE dilution and flow cytometric analysis at 7d. Similar results were seen in two independent experiments with n=3 donors. *** $p<0.0005$, 2-tailed Student's t test. CIITA, class II MHC Transactivator; HECFC, human endothelial colony forming cell; T_{EM}, effector memory T cell.

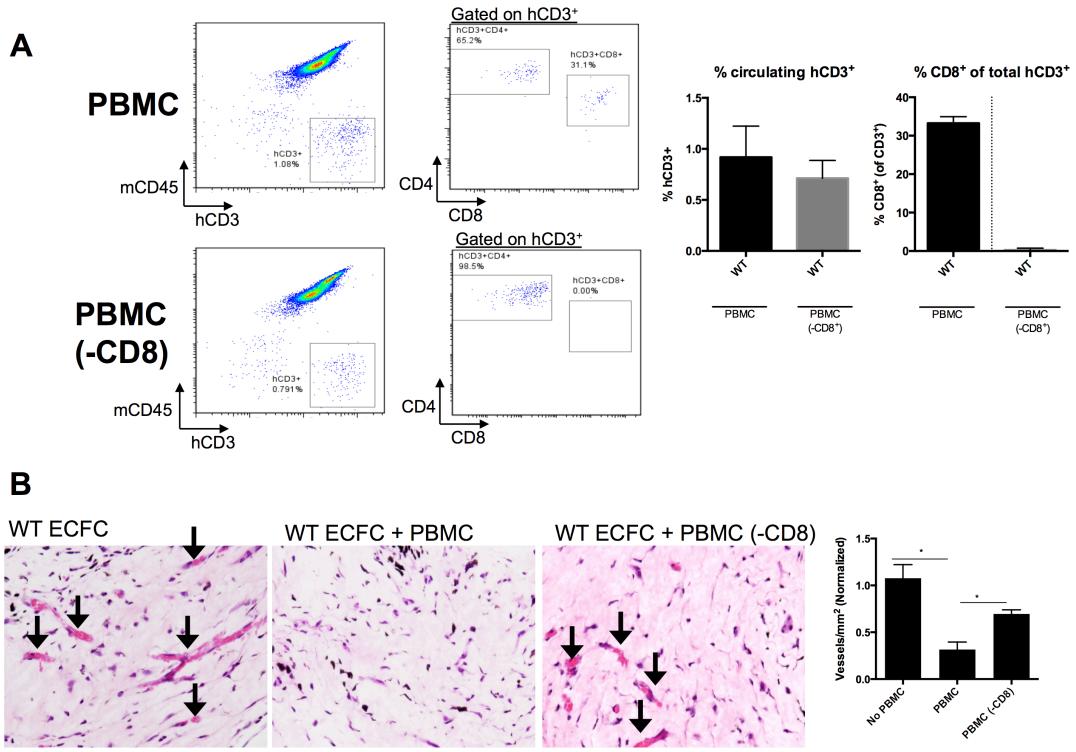

Figure S6: **Depletion of CD8$^+$ cells from PBMC inoculum prior to adoptive transfer results in absence of CD8$^+$ T cells in the peripheral blood and reduces the extent of allogeneic human EC microvessel destruction. (A)** Analysis of circulating human CD3$^+$ subsets from blood taken 10d after adoptive transfer of 2x10^8 human PBMC. As quantified by flow cytometry, pre-depletion of CD8$^+$ effectively prevents appearance of CD8$^+$ cells in the peripheral circulation (n=3). **(B)** Adoptive transfer of whole PBMC into animals bearing synthetic human microvessels results loss of perfused vessels at 10d, whereas removal of CD8$^+$ from the PBMC inoculum significantly inhibits vessel loss (n=3). Similar results were seen in two independent experiments. * $p<0.05$, 2-tailed Student's t test. PBMC, peripheral blood mononuclear cells.