Figure S2. Differential anti-oxidant capacity in human T cell subsets. (A) Human TIL1383I TCR-transduced T cells were kept in different concentrations of H₂O₂ and analyzed for cell death between CD8⁺ CD62L⁺/CD62L⁻ T cell subsets by staining with Annexin V and 7AAD. Experiments were repeated thrice with similar results. (B) PBMCs labeled with CFSE and stimulated with anti-CD3 for five days were used to determine the cell surface thiol expression (c-SH) by flow cytometry using ALM-633 staining. The left panel shows CFSE dilution and the right panel shows the level of thiols in cells that were gated on CFSE⁻ and CFSE⁺ CD8⁺ T cells (indicated by arrows). (C) Unstimulated (-) and stimulated (+) CD8⁺ T cell lysates were prepared and probed for the expression of TRX-1 and TRX-2. Blots were also probed with GAPDH for loading control (Left panel). Bar graphs on the right show densitometry analysis of blots from three similar experiments. Experiment was repeated twice with similar results. (D) GSH/GSSG ratio were measured in FACS sorted CD62L⁻ and CD62L⁺ CD8⁺ T cells. Figure represents cumulative data from three different experiments. (E) Antioxidant array consisting of 84 antioxidant and redox signaling pathway genes was analyzed using real time PCR in 2 different samples from FACS sorted human CD62L⁻ and CD62L⁺ CD8⁺ T cells from normal healthy donors. (F) Human gp100 reactive CD8⁺ T cells from pMel mouse were TCR-activated with cognate antigen along with IL-2 for 3 days and sorted on the basis of c-SH expression into c-SH⁺ or c-SH⁻ fractions. Sorting strategy is shown.
Figure S2