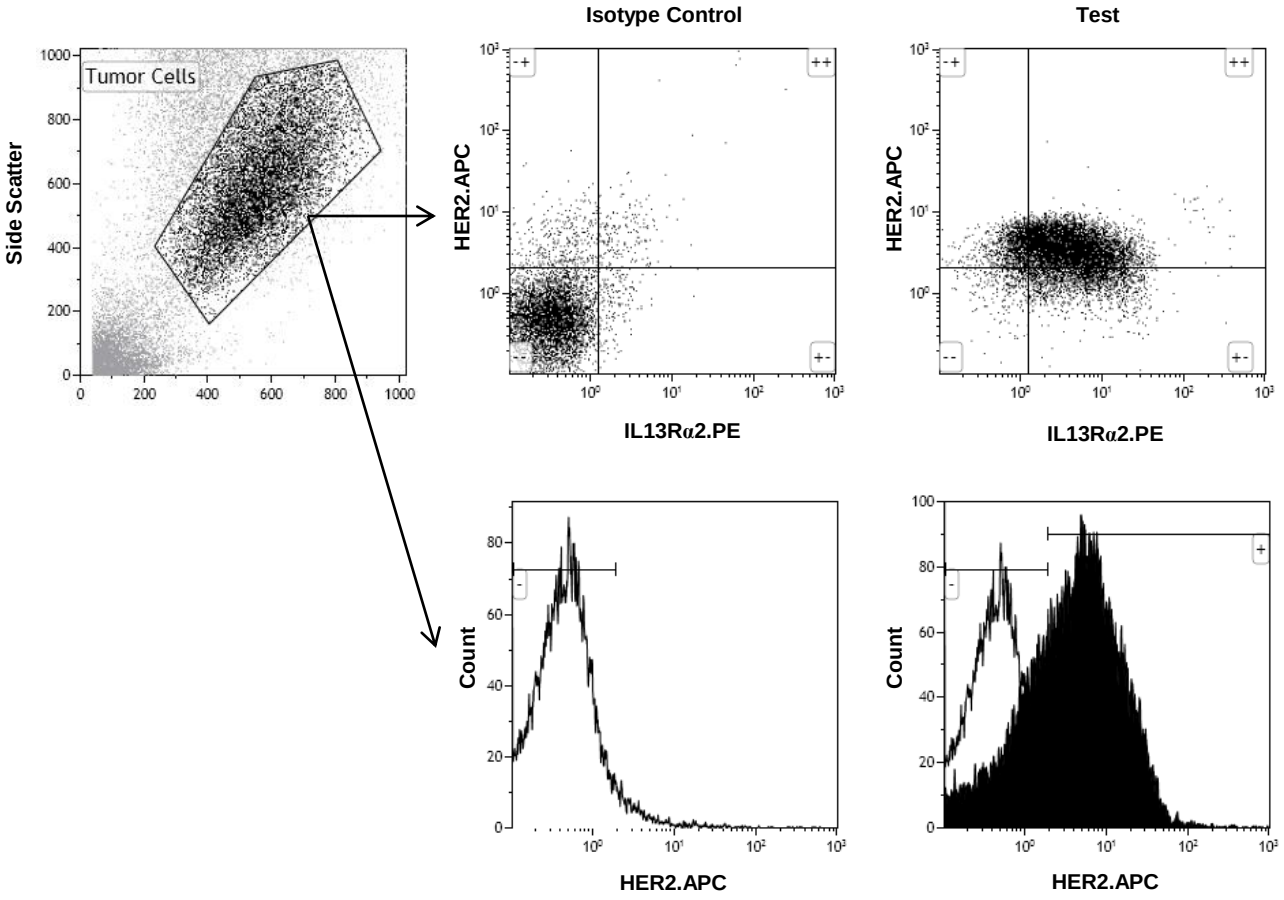
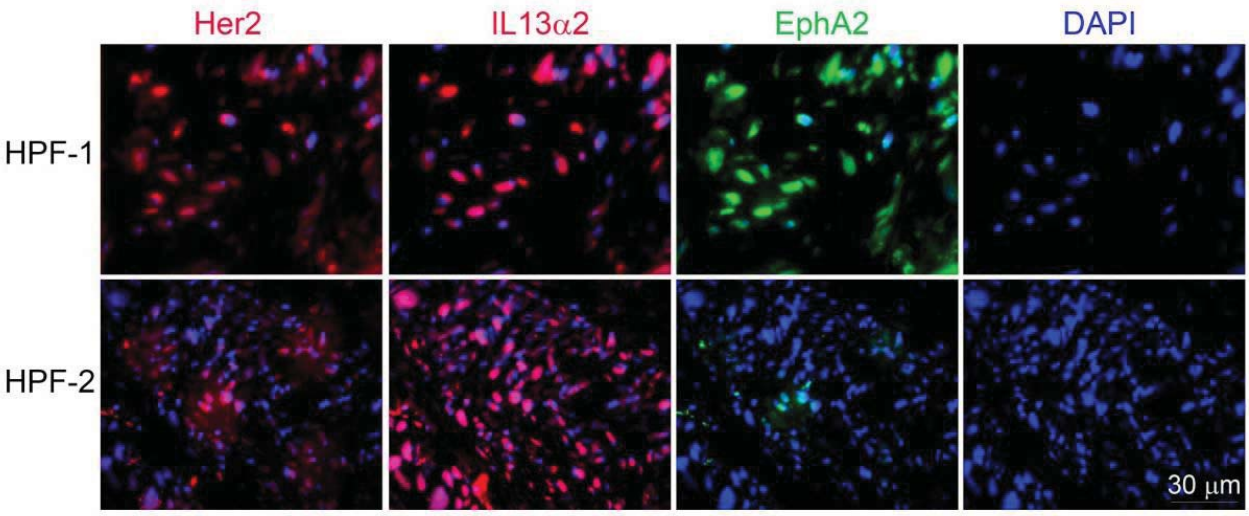
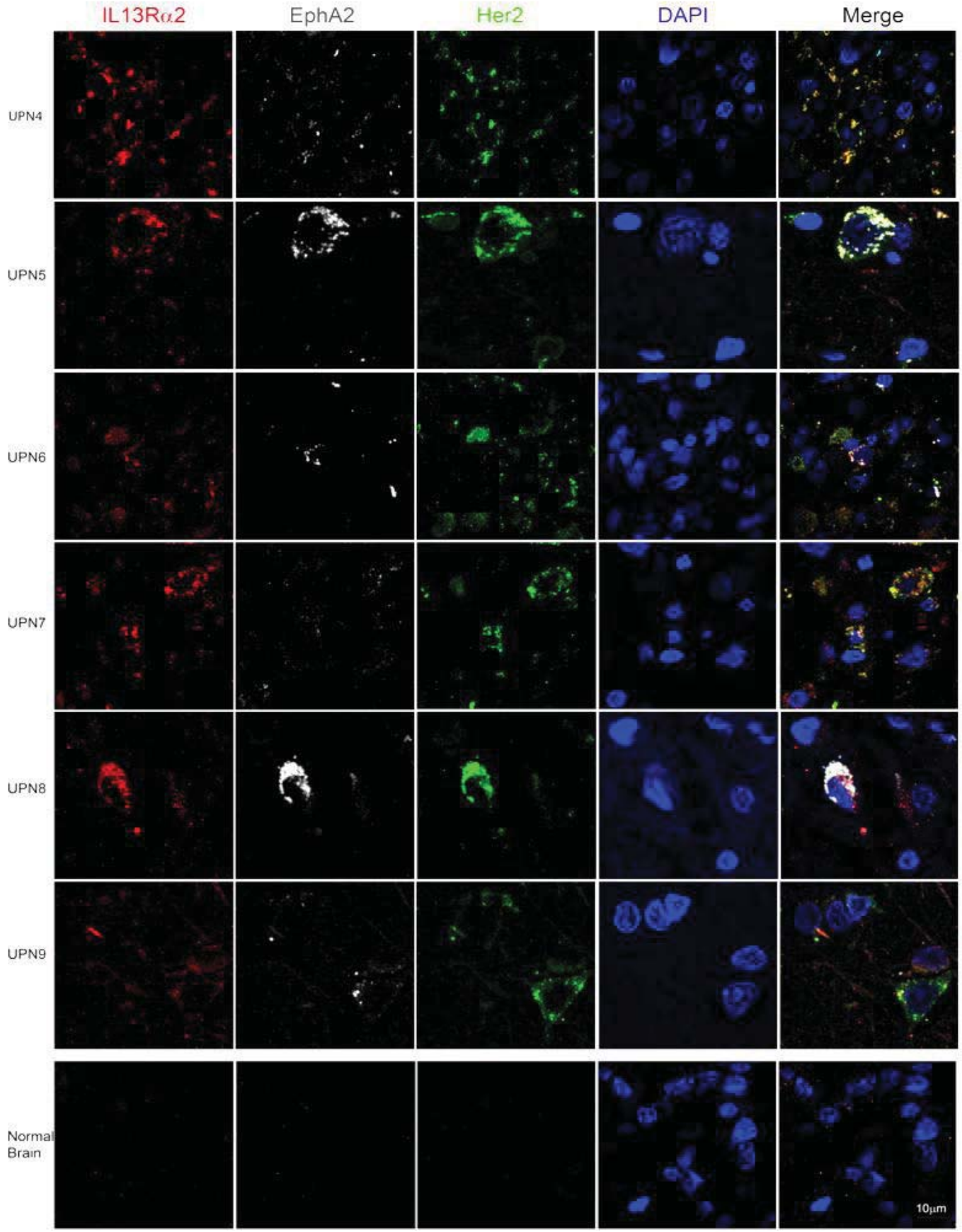


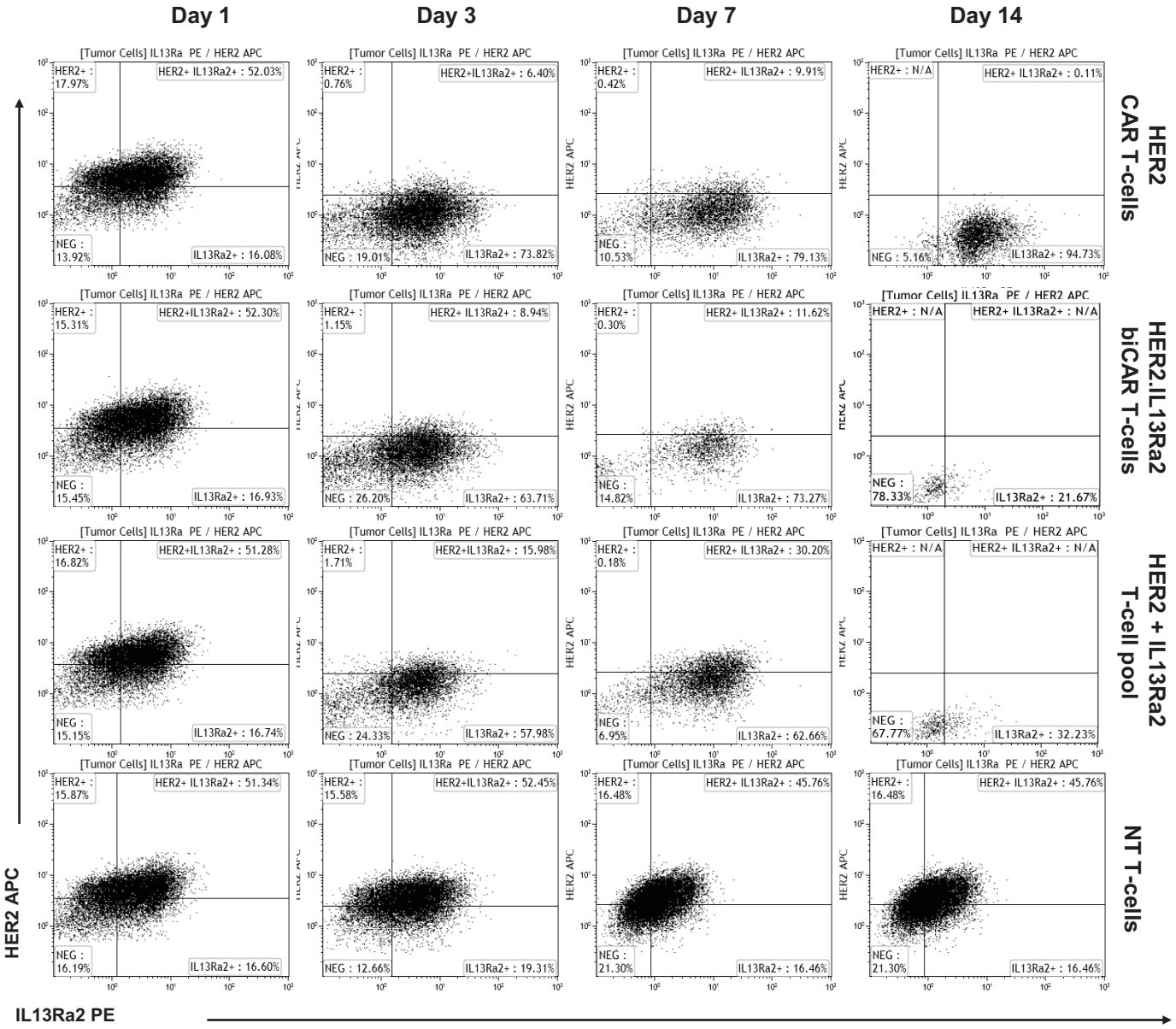
Supplemental Figure 1: Gating strategy for tumor cell analysis performed in Figures 1 and 2 and Supplemental Figure 4.



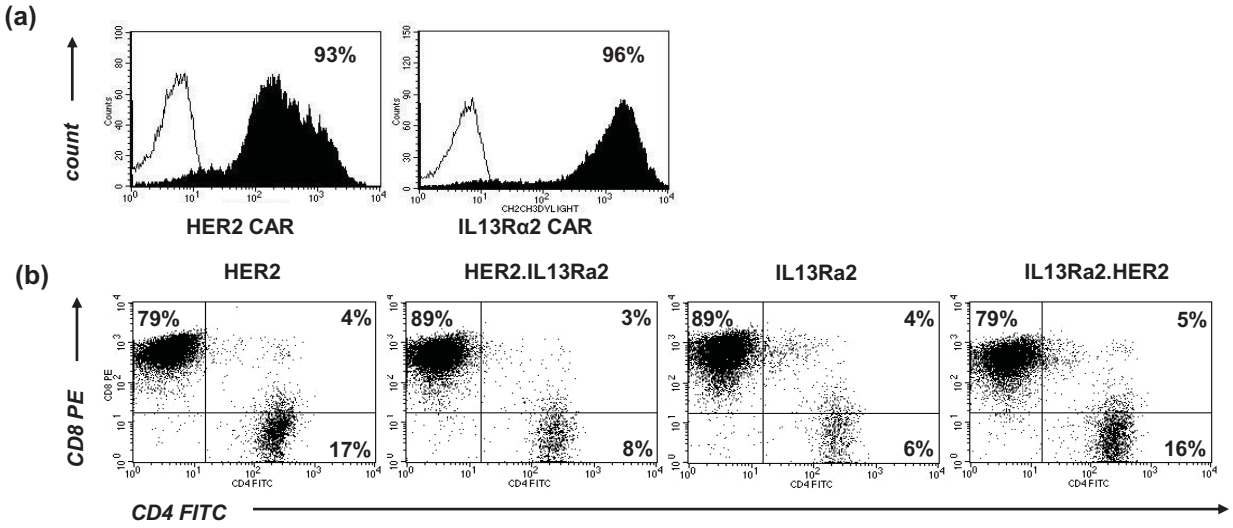
Supplemental Figure 2: Co-immunofluorescence for HER2, IL13Rα2 and EphA2 demonstrating the heterogenous expression of antigens within the same tumor by changing the field examined. HPF=high power field.



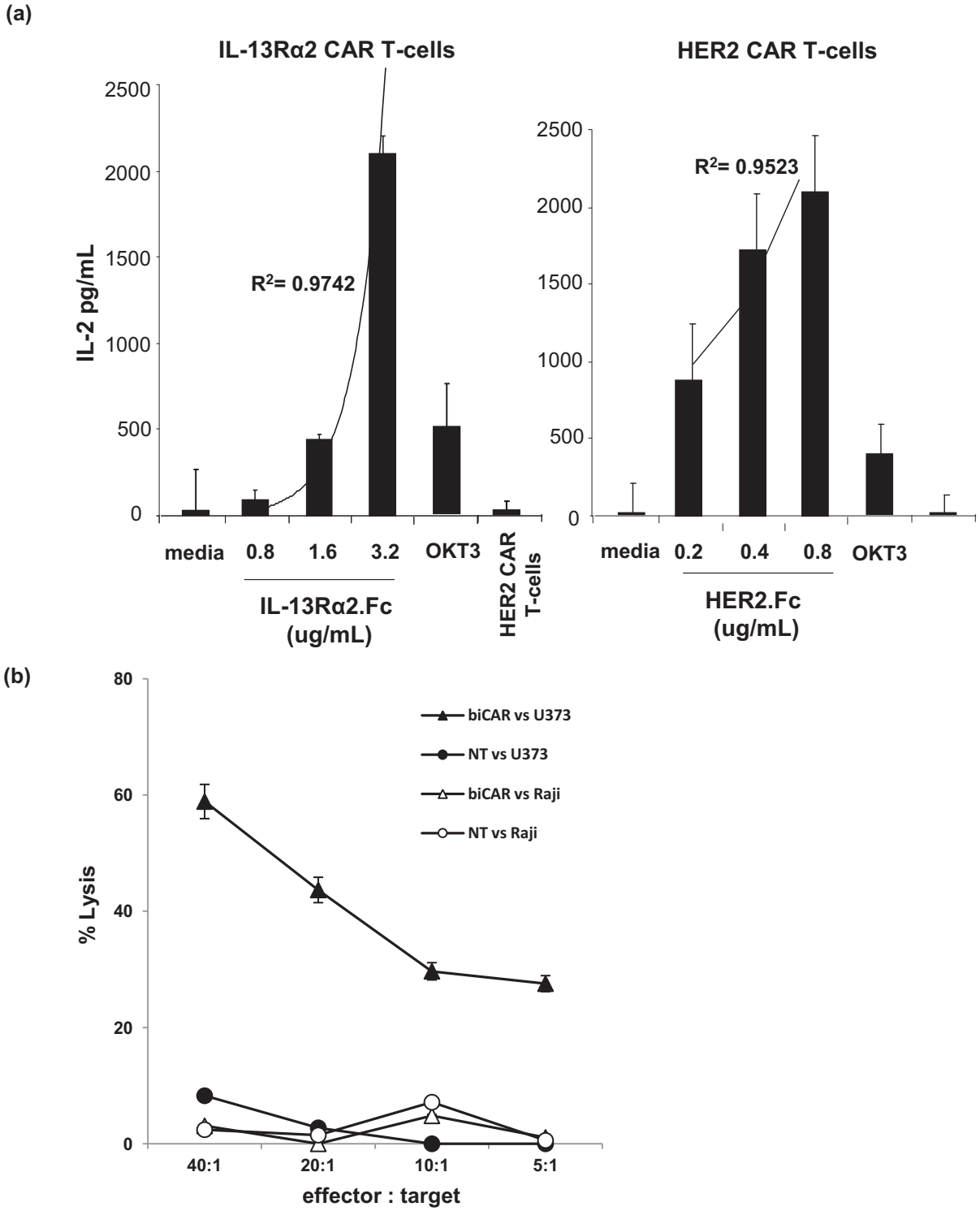
Supplemental Figure 3: Representative co-immunofluorescence captures for HER2, IL13Rα2 and EphA2 from 6 serially diagnosed GBM patients and normal brain tissue. Figure 3 depicts the quantification of this data using Image J® software.



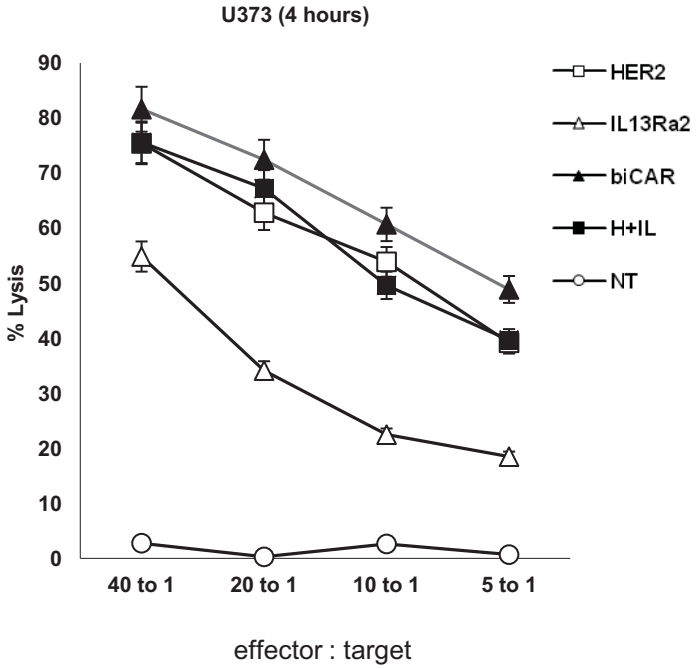
Supplemental Figure 4: Flowcytometric analysis of IL13Rα2 and HER2 expression in the glioma cell line U373-GBM upon encounter of HER2 CAR T cells, IL13Rα2 CAR T cells, their pooled product and biCAR T cells at 1, 3, 7 and 14 days of coculture. NT (non transduced T cells) are control.



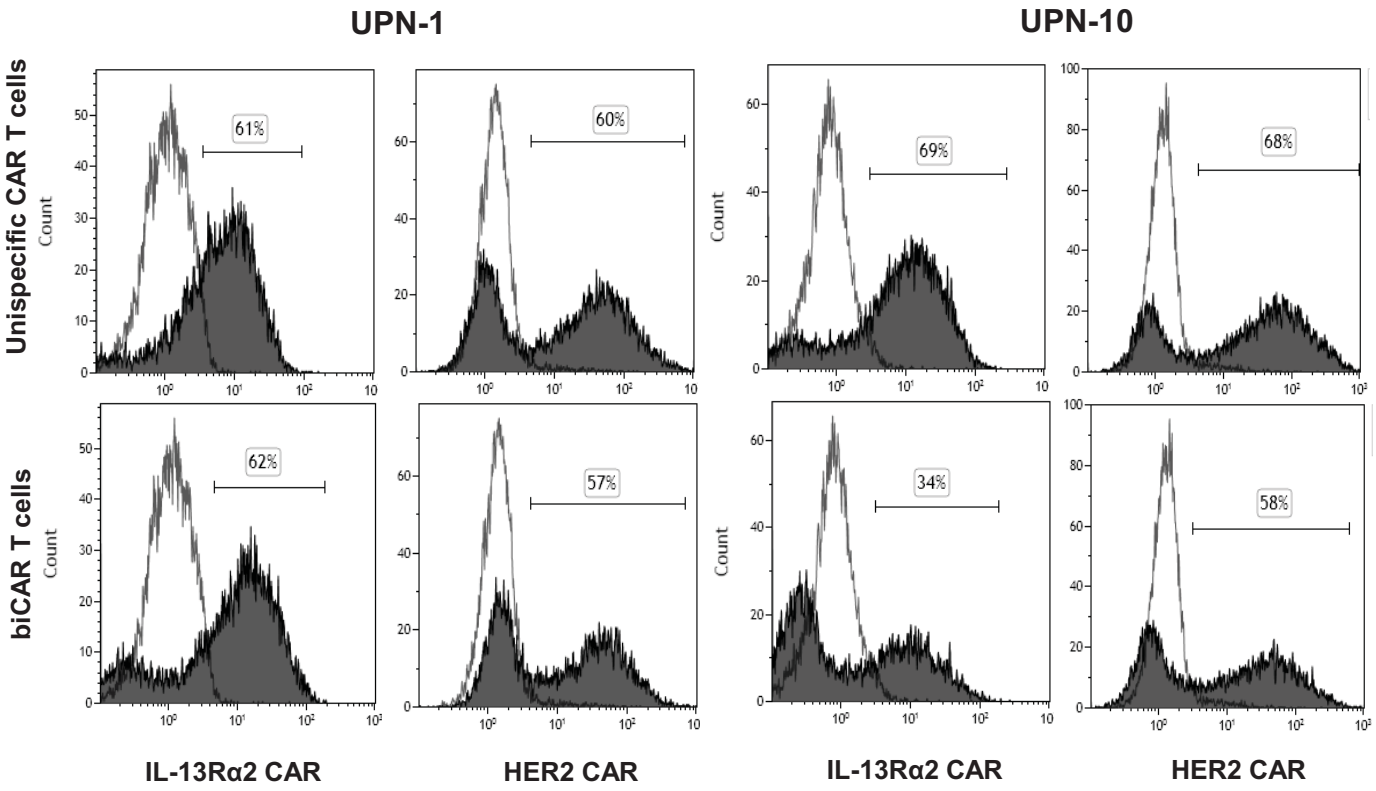
Supplemental Figure 5: (a) Transduction efficiencies of biCAR T cells and (b) their CD4/CD8 phenotype.



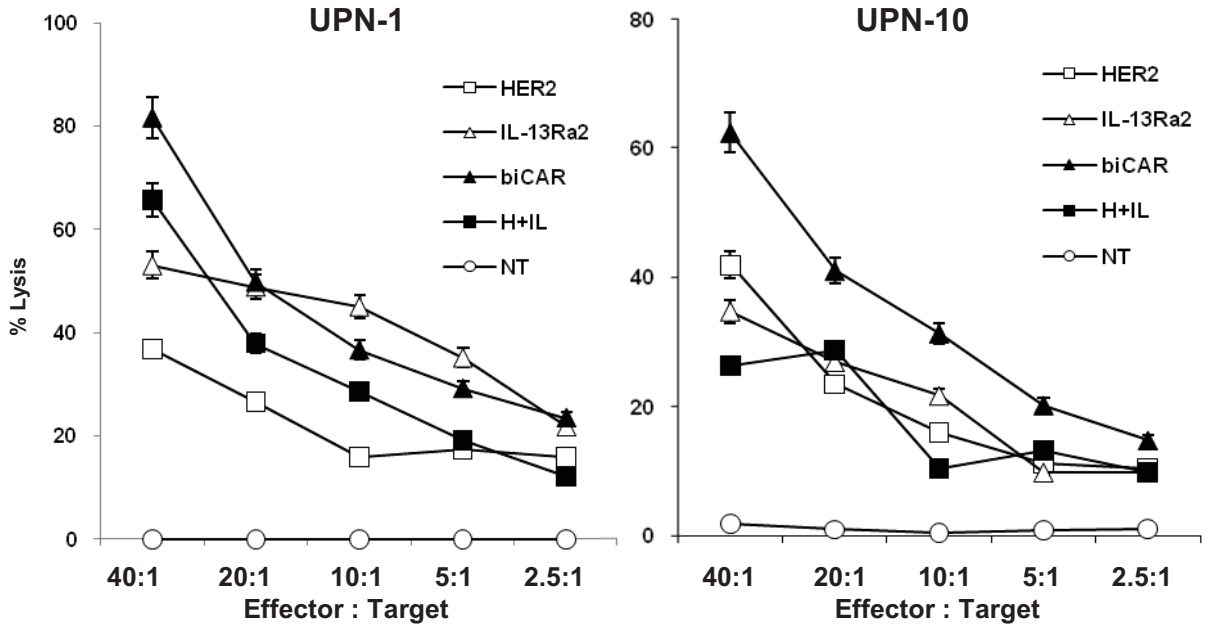
Supplemental Figure 6: (a) validation of the distinct reactivity of T cells to plate bound IL13Rα2-Fc and HER2-Fc chimeric proteins. Both HER2 CAR T cells and IL13Rα2 CAR T cells reacted proportionately to their respective antigens but not to a mock coating of the opposite antigen. Stimulation experiments in Figure 5c were performed using protein concentration in the validated rance. (b) biCAR T cells recognized and killed the HER2/IL13Rα2-expressing U373-GBM but failed to lyse Raji, the HER2/IL13Rα2-null Burkitt's lymphoma line. NT cells had background lytic activity. Shown is one representative of 3 experiments performed in triplicates.



Supplemental Figure 7: A standard 4 hour ^{51}Cr release cytotoxicity assay of HER2 CAR T cells, IL13Rα2 CAR T cells, their pooled product (HIL) and biCAR T cells against the HER2, IL13Rα2 positive glioma line U373-GBM. Shown is one representative of 3 experiments performed in triplicates.



Supplemental Figure 8: Transduction efficiencies unispecifc CAR T cells and of biCAR T cells generated from two GBM patients, UPN-1 and UPN-10.



Supplemental Figure 9: An extended standard 4 hour ^{51}Cr release cytotoxicity assay of primay GBM patient effectors, namely: HER2 CAR T cells, IL13Rα2 CAR T cells, their pooled product (HIL) and biCAR T cells against autologous glioma cells. Shown is one representative of 2 experiments performed in triplicates.