Supplementary information for:

An oxygen sensitive self-decision making engineered CAR T-cell

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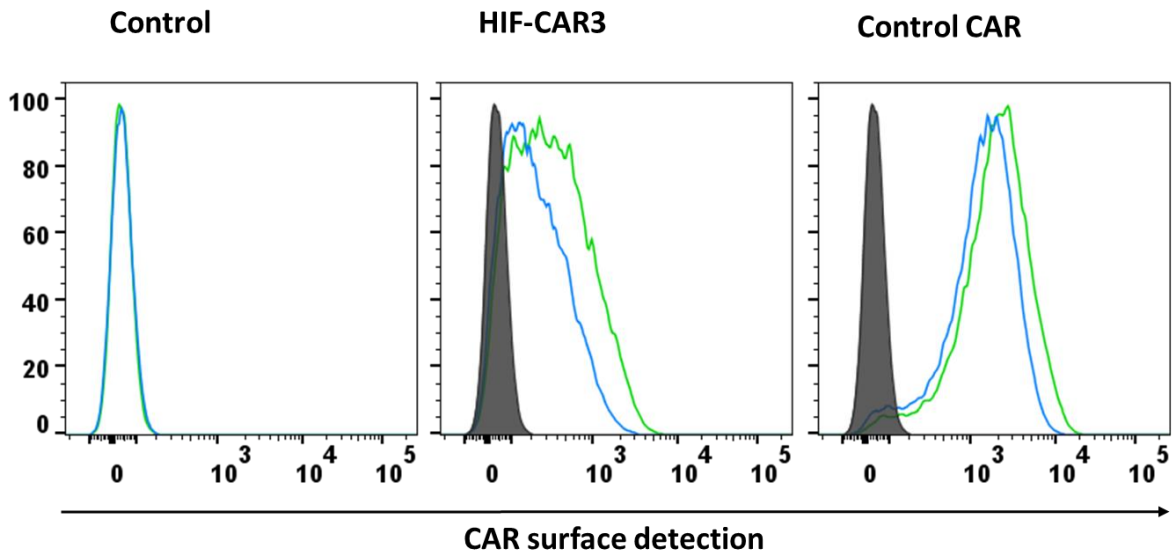
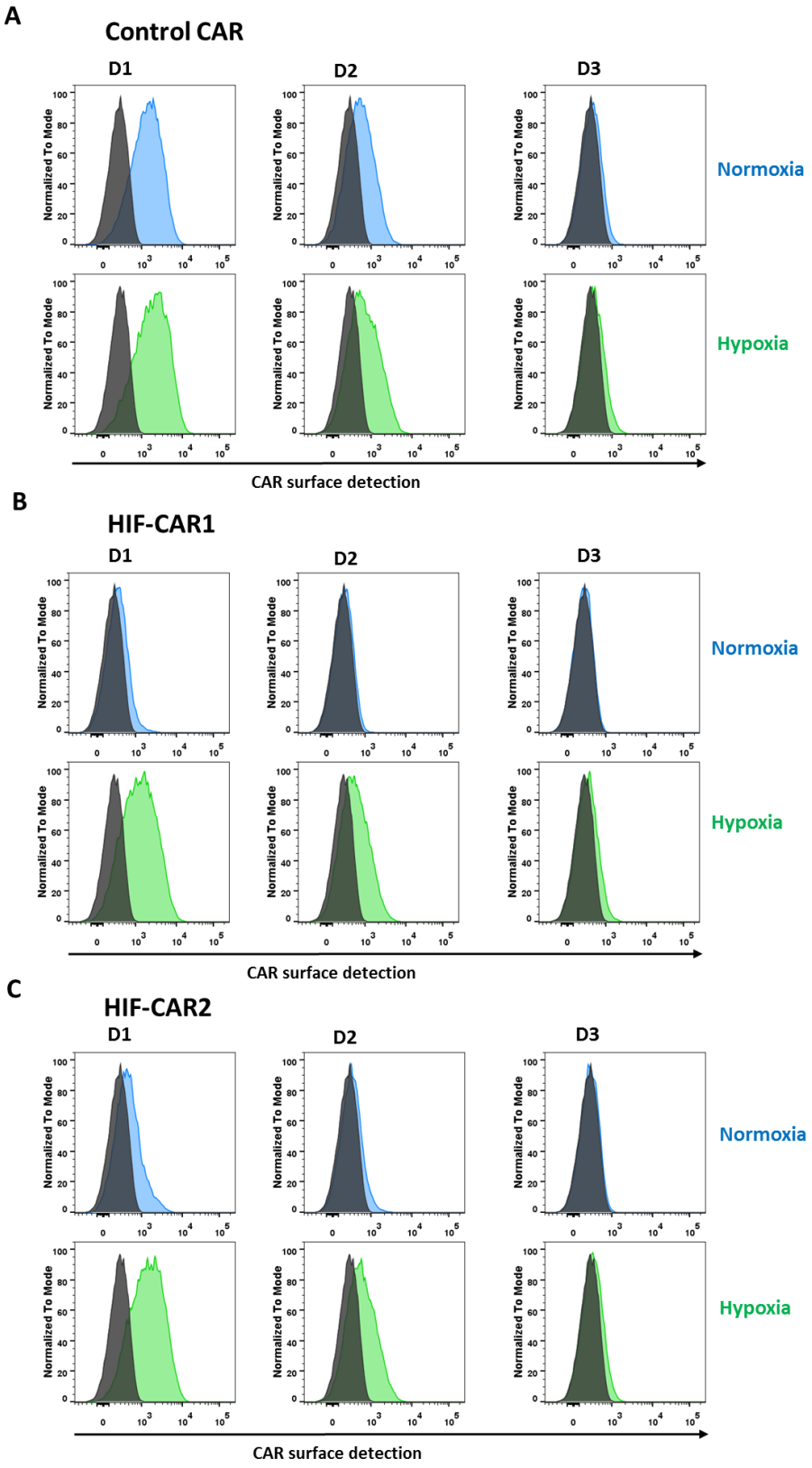
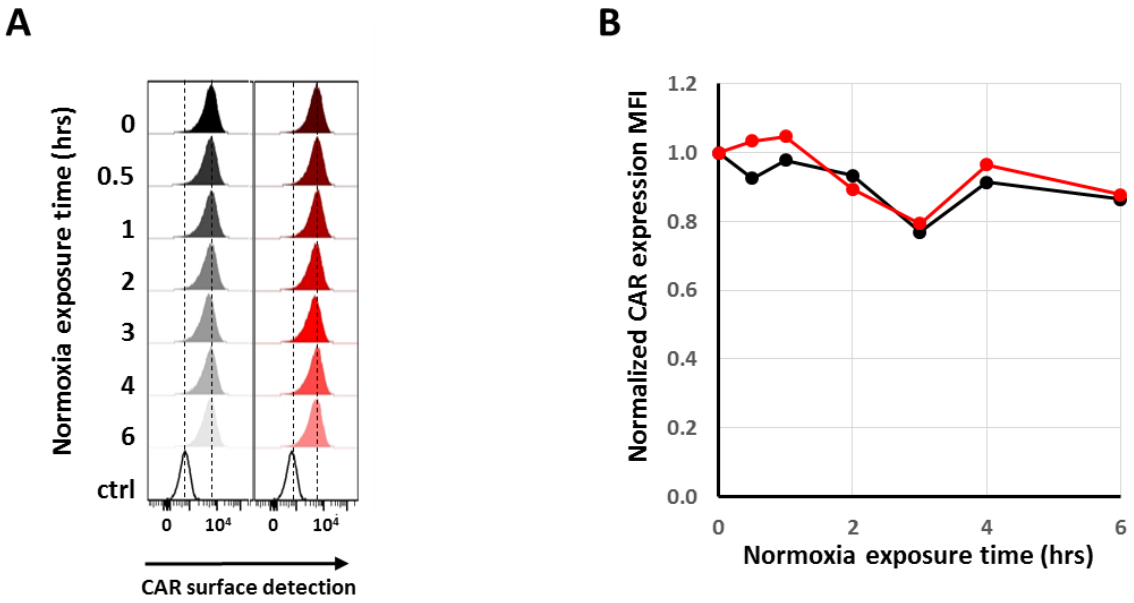


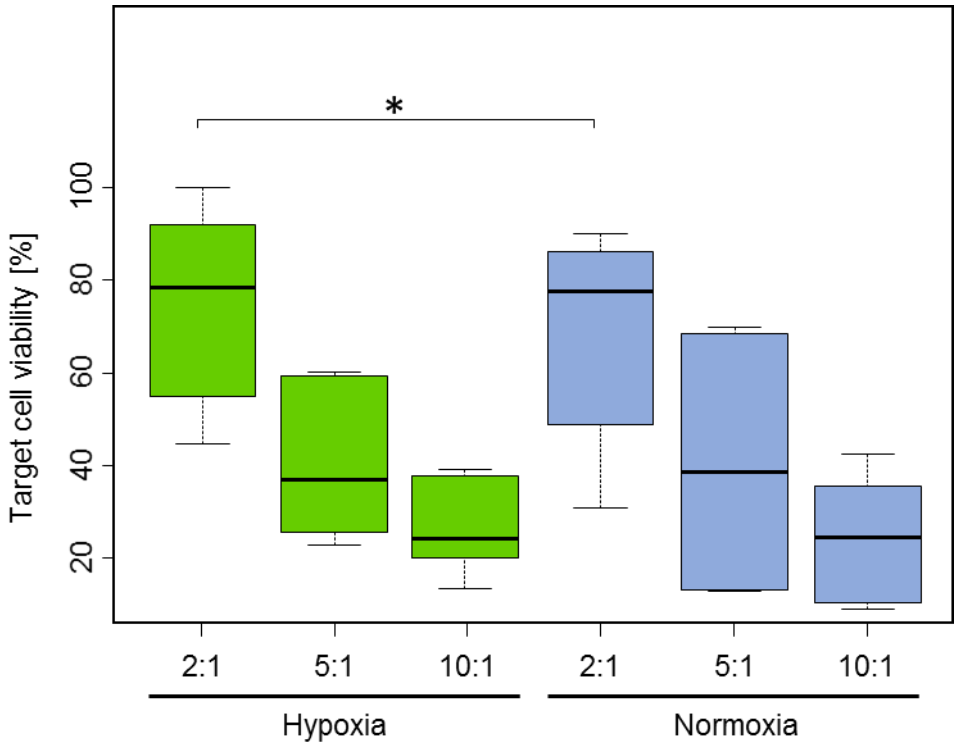
Supplementary figure 1: Impact of the oxygen level on the surface presentation of the engineered HIF-CAR3. FACS histogram representations of the detection of the different CARs (control and HIF-CAR3) under hypoxic or normoxic conditions.



Supplementary figure 2: Impact of the oxygen level on the surface presentation of the engineered CAR. FACS histogram representations of the detection of the different CARs (control, HIF-CAR1 and HIF-CAR2) under hypoxic or normoxic conditions with different doses of transfected mRNA.



Supplementary figure 3: Determination of the CAR surface presentation decay after removal of the hypoxia input. (a) FACS histogram representation of the CAR detection (HIF-CAR3 in red and control CAR in black) at the surface of primary T-cell after removal of the hypoxia input over a 6h period. The detection of the Fab'2 region of the scFv is shown. (b) Time course analysis of CARs (HIF-CAR3 in red and control CAR in black) surface presentation decay after removal of the hypoxia input. The MFI values were normalized to 1 for each CAR at the time at which the normoxic condition were re-established.



Supplementary figure 4: Cytolytic properties of the control CAR T-cells. The effect of the difference of oxygen levels (normoxia and hypoxia) on the cytolytic capacities of the of the CAR T cells toward model antigen presenting cell was assessed in a luciferase-based assay. Boxplots representing the percentage of viable Daudi target cell after coculture with CAR T-cells. E/T=2, E/T=5, E/T=10. E/T denote the effector/target ratios, N=6 with each experiments done in three technical replicates. Significant differences in viability between normoxic and hypoxic conditions are indicated. Significance is determine by a standard paired t-test, * = $p \leq 0.05$.