



Supplementary Figure 2. Impact of spacer region on T cells differentiation

(A) Memory phenotype of the CAR T cells was defined analysing CD62L and CD45RA expression, by FACS analysis, at day10. T_{SCM} (CD45RA+/CD62L+ memory stem T cells), T_{CM} (CD45RA-/CD62L+ central memory T cells), T_{EM} (CD45RA-/CD62L- effector memory T cells), T_{EMRA} (CD45+/CD62L- effector memory RA T cells). CD19-NWL CAR T cells were used as a control. Memory phenotype of the different CAR T cells from a representative donor is shown. Paired t-test on SCM was statistically significant for untransduced (UNT; =0.01; n=7), NWN2 (P =0.01; n=6), NWS (P=0.01; n=4), and NMS (P = 0.001; n=7), compared to NWL. (B) FACS analysis (n=3) showing the exhaustion status of CAR T cells endowed with the NWL or NWN2 spacers; PD1, TIM3 and LAG3 are exhaustion markers.