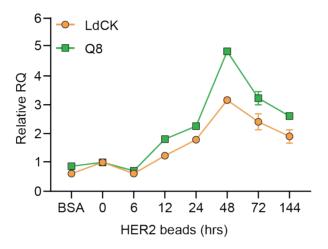


B Kinetics of LdCK-Q8 expression



RB-340-1 CAR-T manufacture workflow.

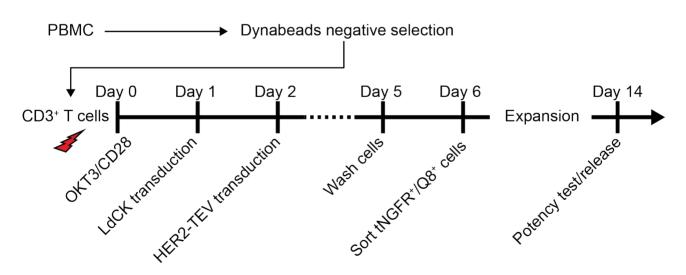


Figure S1, related to Figure 1.

C

(A) Comparative assessment of expression of HER2 CAR according to the anti-4D5 epitope idiotype antibody recognizing HER2 directly compared to the detection of the tNGFR tag. The latter is a preffered method for sorting and phenotyping of cells since it has no direct biological effect on T cell activation/function.
(B) Kinetics of expression of dCas9 and Q8 by qPCR. (C) CAR-T manufacture process – CD3+ T cells are negatively selected from peripheral blood monocytes and stimulated with OKT3/CD28. LdCK is added for transduction a day later followed on the second day by HER2-TEV. Five days after the original stimulation the activation is removed and on the following day cells are sorted to enrich the tNGFR+/Q8+ population.

The expansion is then continued till time of release between day 14 of production. Cells are also tested at that point for permanence of transduction as shown in *Figure 1A*.