

Webinar outline

- Discovery of 4-1BB and 4-1BB immuno-biology
 - **Byoung S. Kwon, PhD** - *Eutilex*
- 4-1BB as an important prosurvival signal for CAR T cells
 - **Michael Milone, MD, PhD** - *University of Pennsylvania*
- Novel antibody-based approaches targeting the 4-1BB pathway
 - **Christian Klein, PhD** - *Roche Innovation Center Zurich*
- Translational and reverse translational research in 4-1BB co-stimulation
 - **Ignacio Melero, MD, PhD** - *CIMA, Clinica Universidad de Navarra*

4-1BB as an important prosurvival signal for CAR T cells

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SITC 4-1BB Webinar, Oct 21, 2021

Disclosure Information

Michael C. Milone

I have the following financial relationships to disclose:

Consultant for:

Speaker's Bureau for:

Grant/Research support and royalties / IPR from: *Novartis, Cabaletta Bio, Verismo Therapeutics*

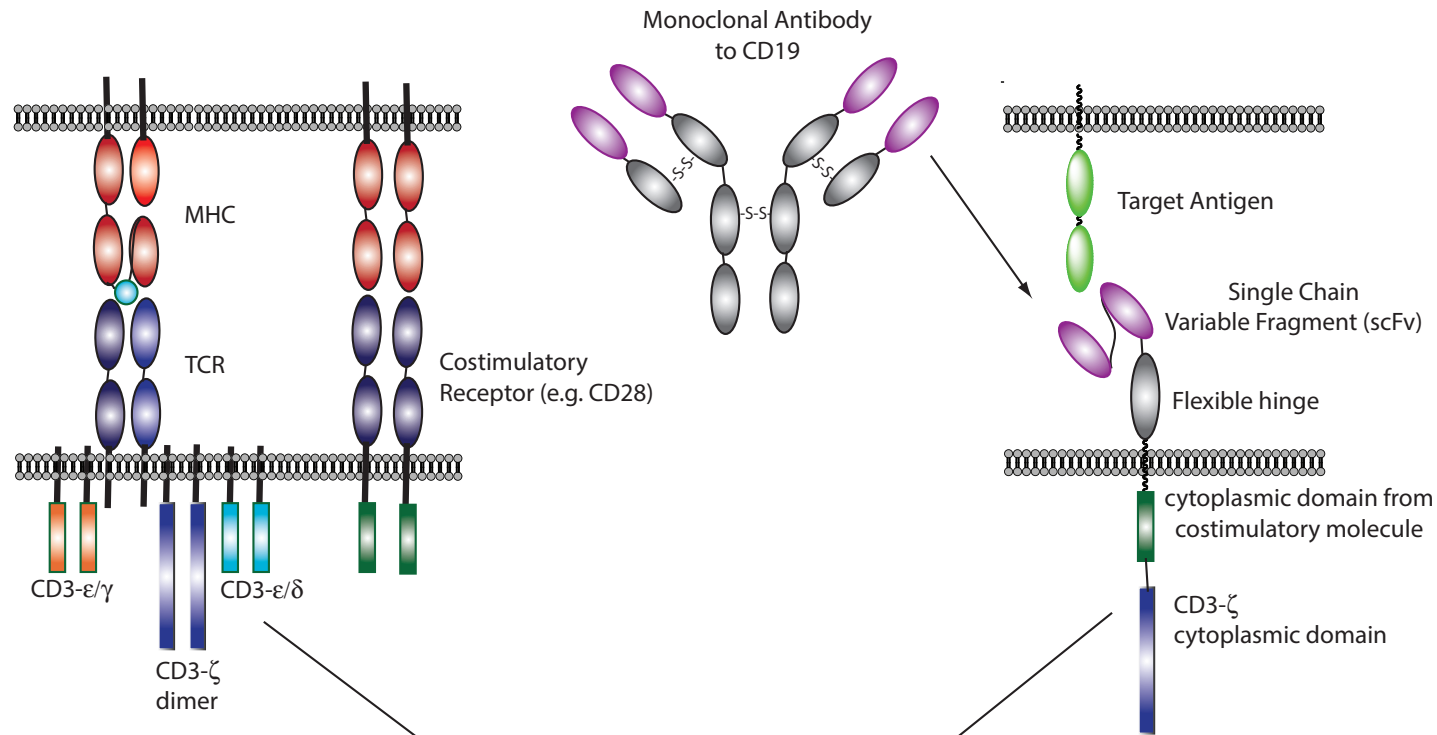
Stockholder in: Cabaletta Bio (CABA), Verismo Therapeutics

Honoraria from:

Employee of:

I am inventor on several patents related to CAR T cell technology that have been licensed and I receive royalties related to this IP

Chimeric Antigen Receptors (CARs): A Synthetic Approach to Engineering Immunity

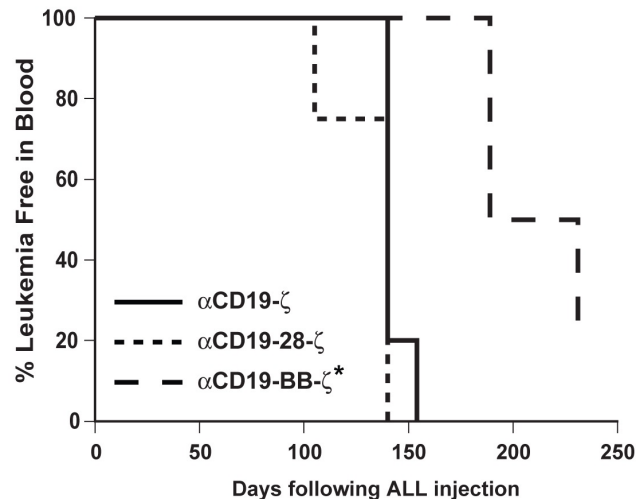
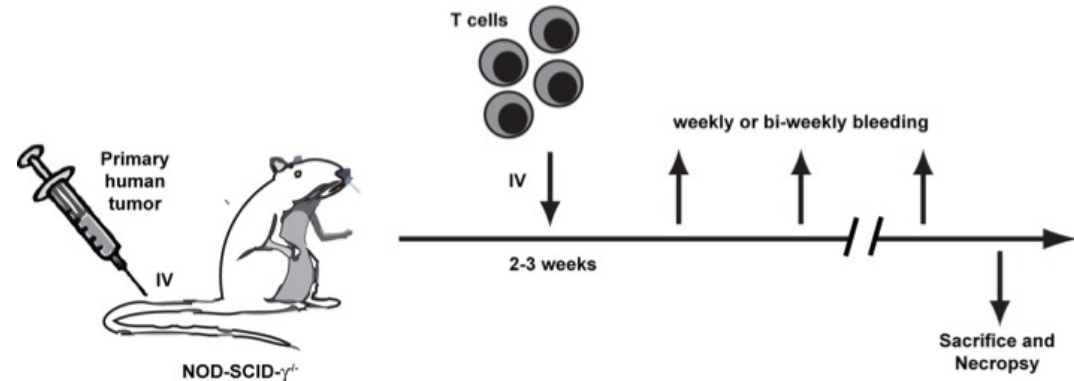
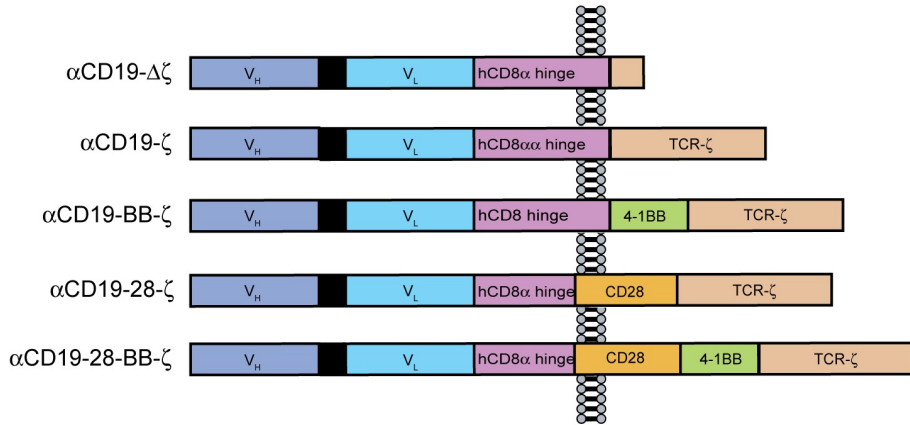


TCR approach can target most proteins expressed by cell

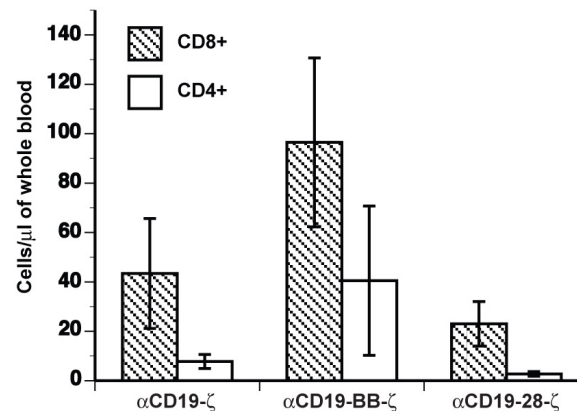
CAR approach is limited to targets (protein and non-protein) expressed on the cell surface

What is the optimal costimulatory domain?

CARs with a 4-1BB cytoplasmic domain exhibit greater anti-leukemic efficacy in vivo



3 weeks post-T cell infusion



4-1BB costimulated CAR T cells exhibit:

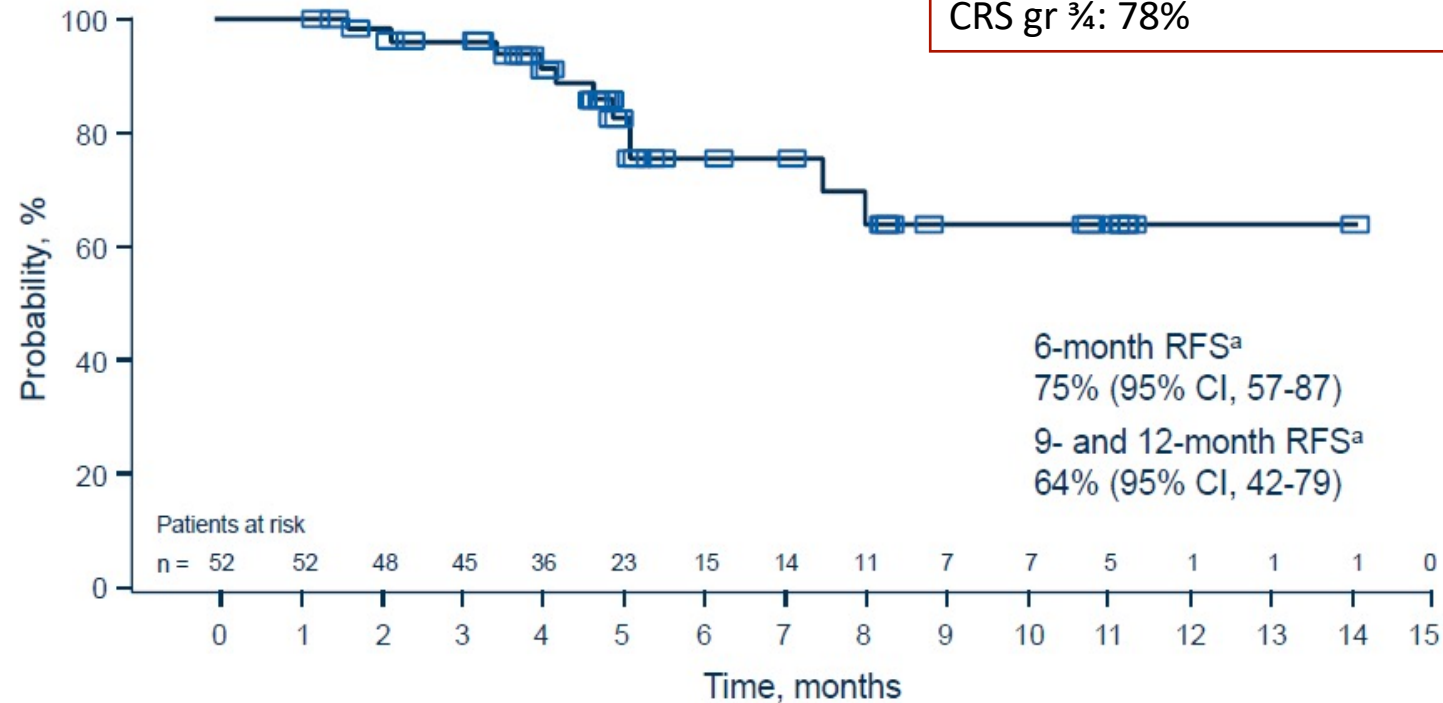
- the greatest anti-leukemic activity
- Activity is associated with persistence following transfer

Tisagenlecleucel in R/R pediatric and young adult ALL

ELIANA study (NCT02435849)

Duration of Remission

CR rate: **83%** (52/63)
All CRs were **MRD-neg**
CRS gr ≥ 3 : 78%



6-month RFS^a
75% (95% CI, 57-87)
9- and 12-month RFS^a
64% (95% CI, 42-79)

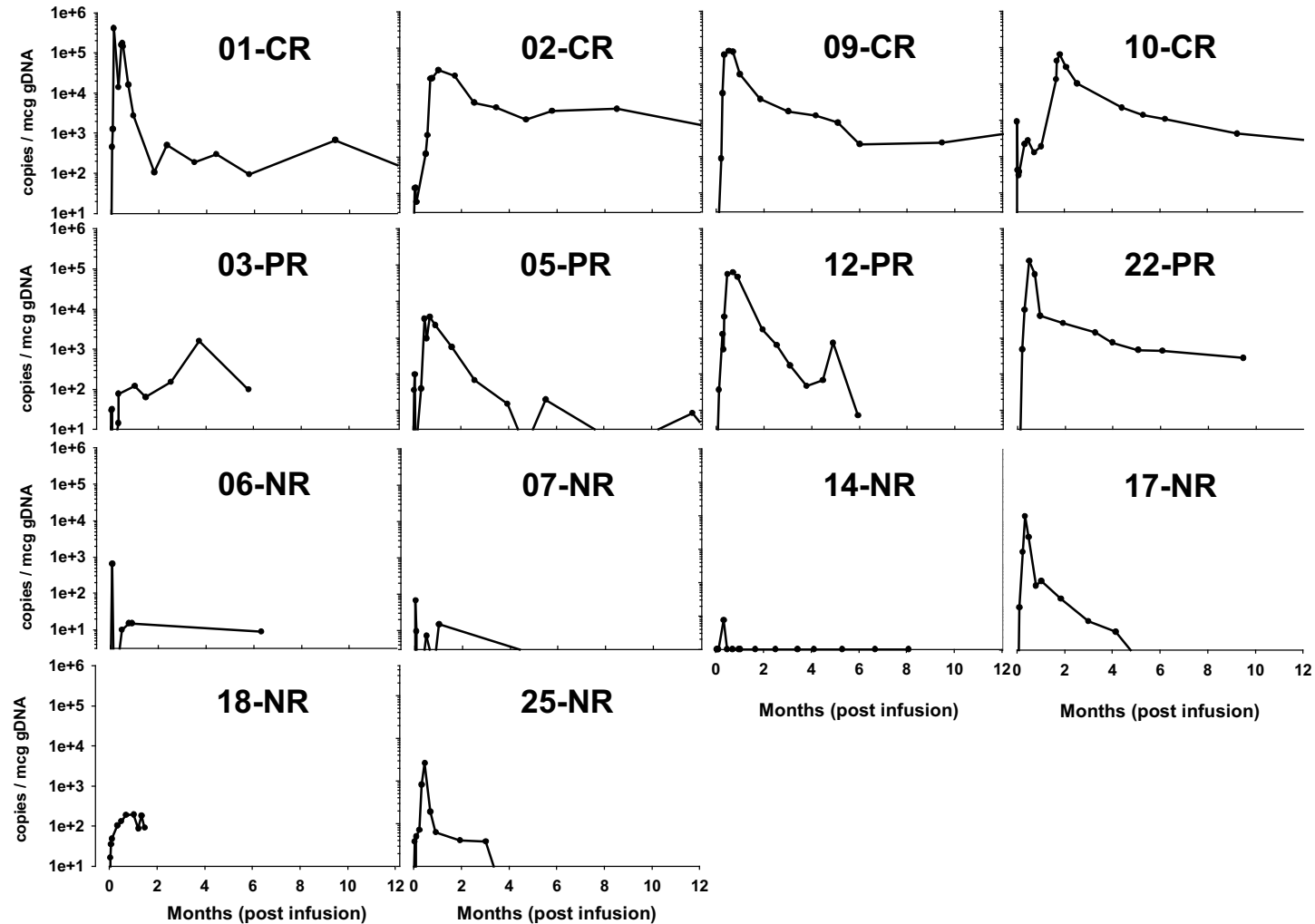
Patients (N = 52)

Number of events (n = 11)

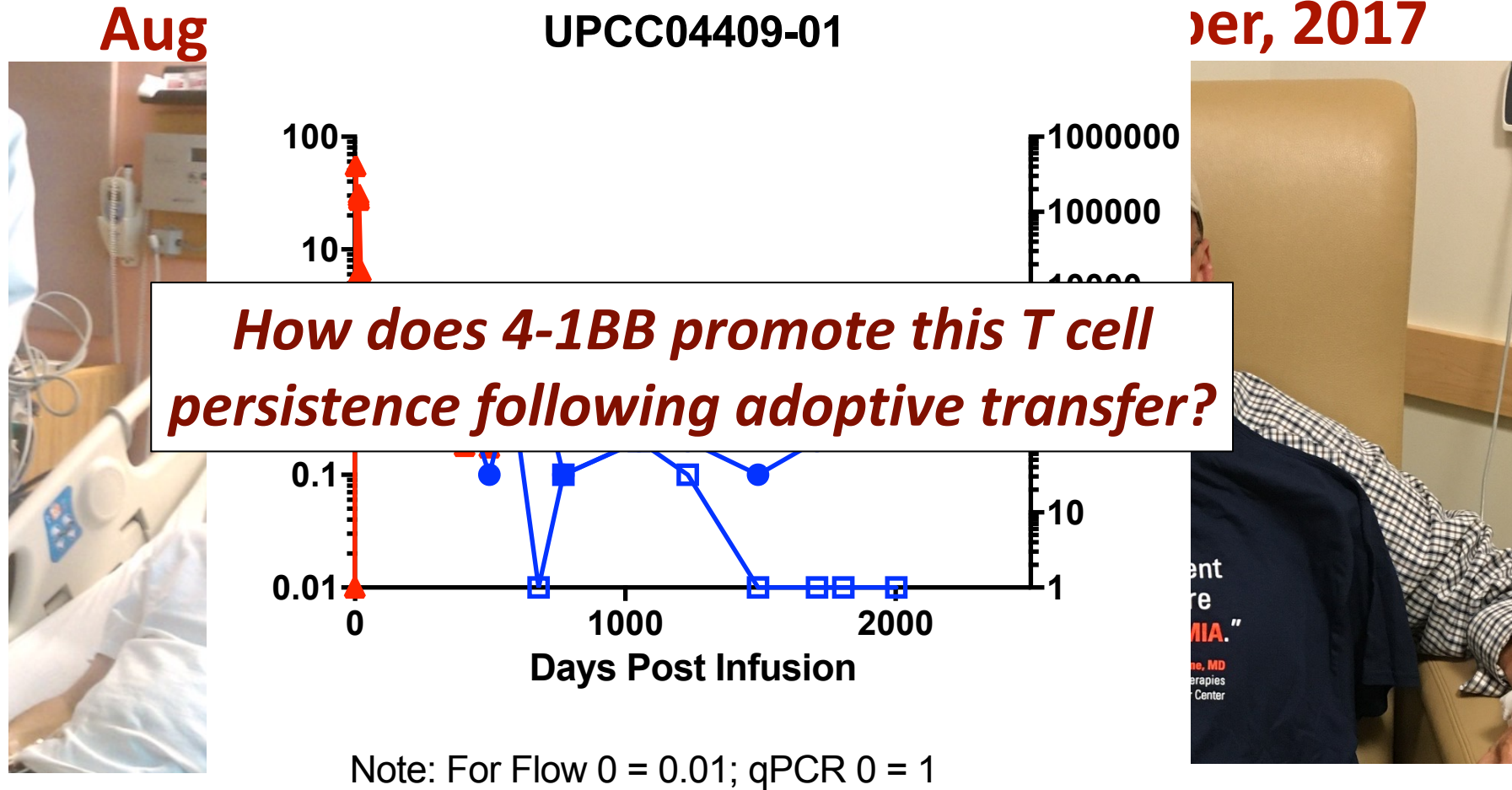
Median follow-up, 4.8 months
Median DOR, not reached

Only patients who achieved CR or CRi were included. Time is relative to onset of remission.
^a Efficacy analysis set.

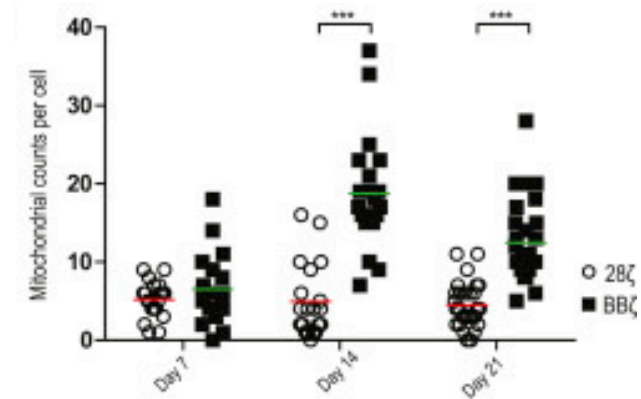
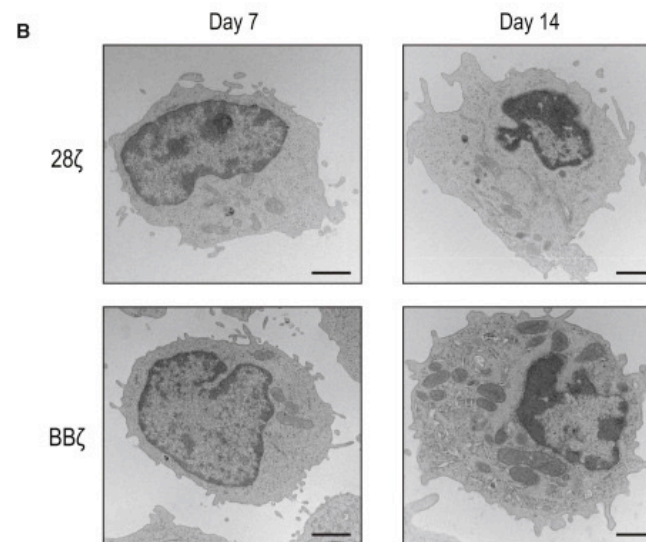
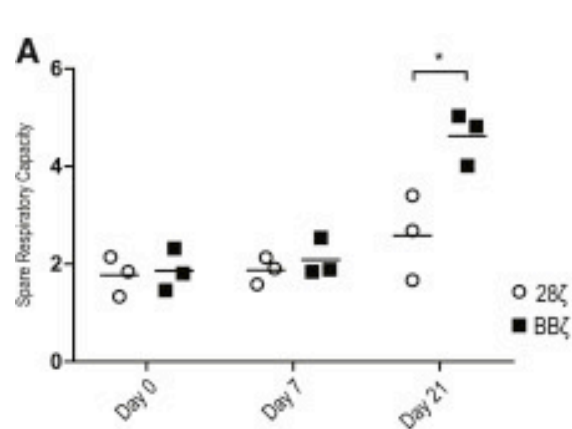
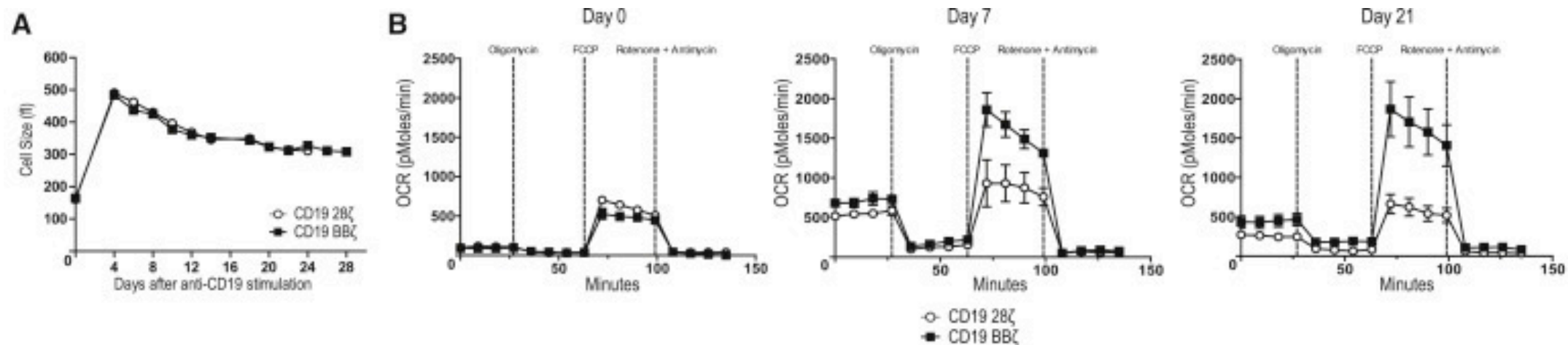
Long term persistence and expression of CTL019 in CLL patients with durable remission



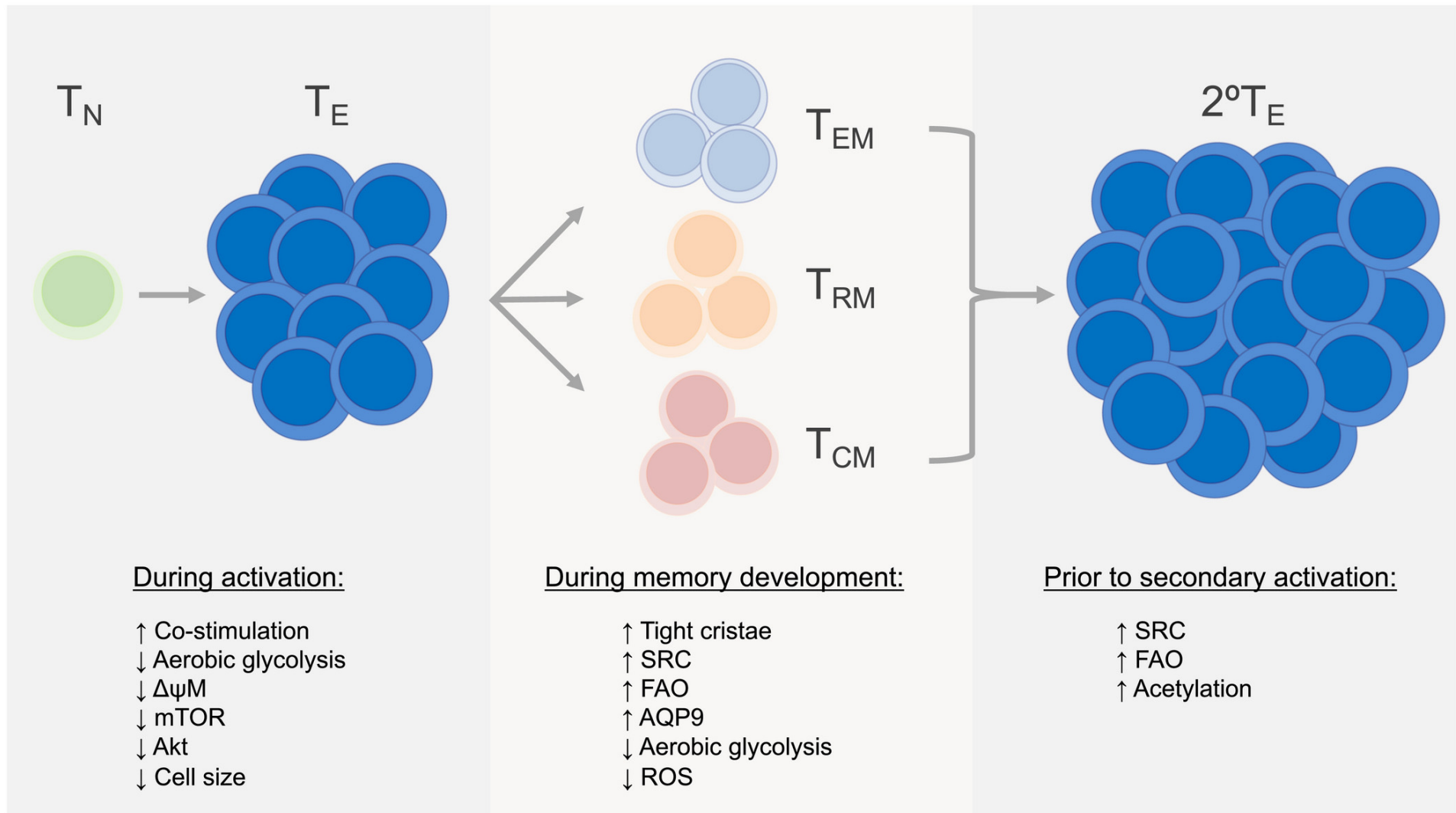
First patient treated with CART19 at Penn had CLL



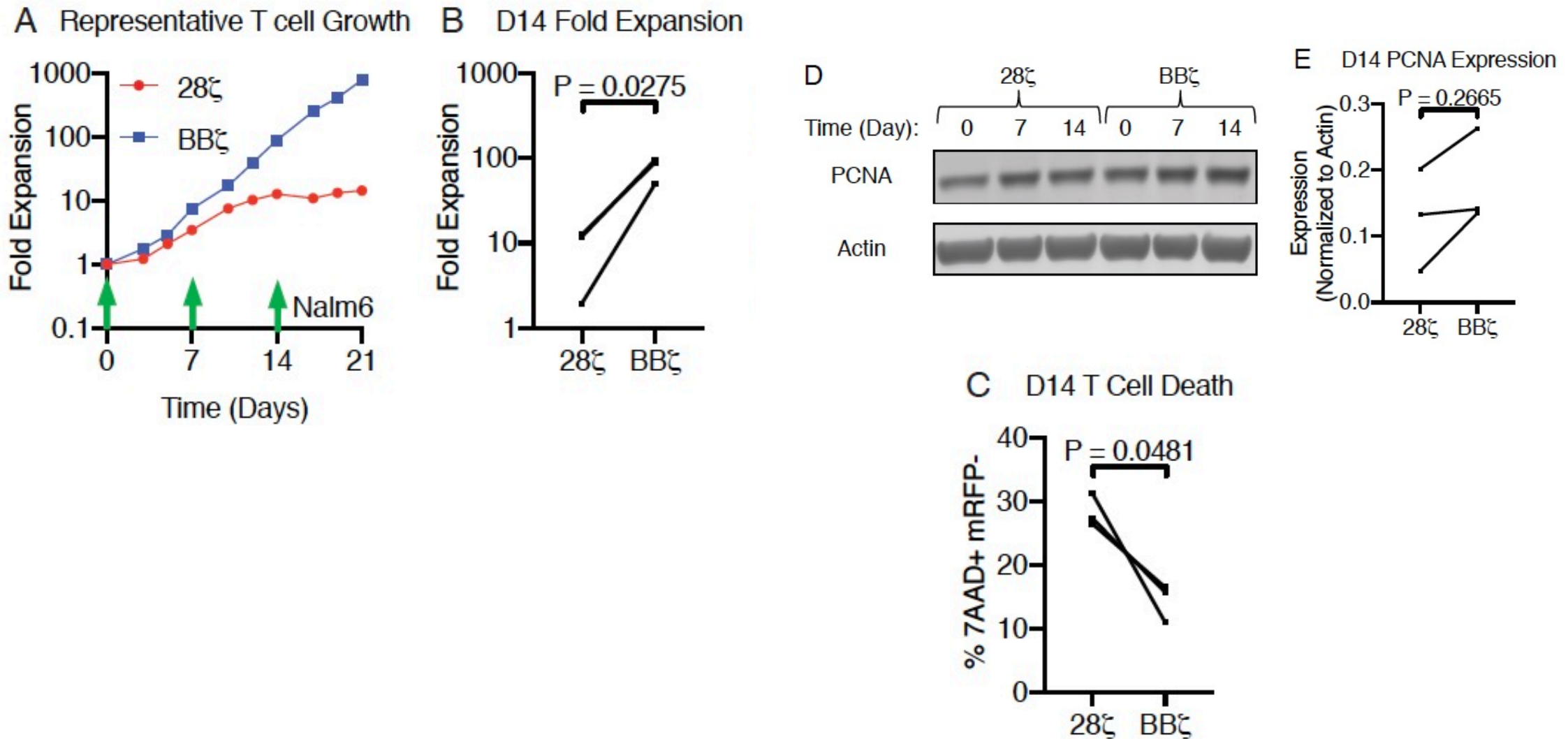
4-1BB promotes development of T cells with a more oxidative metabolic phenotype



T cell memory and metabolism are linked



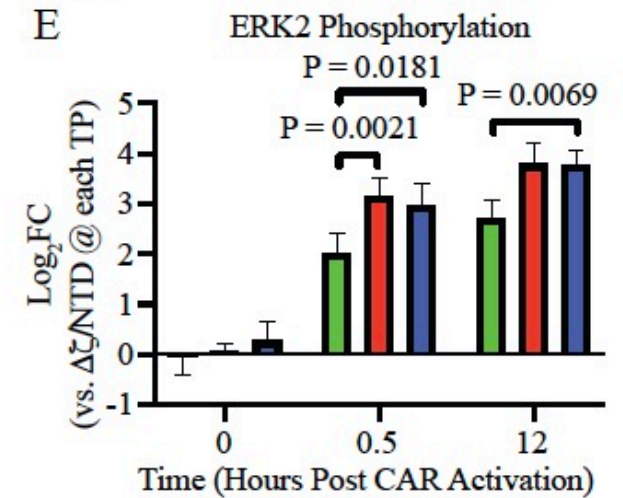
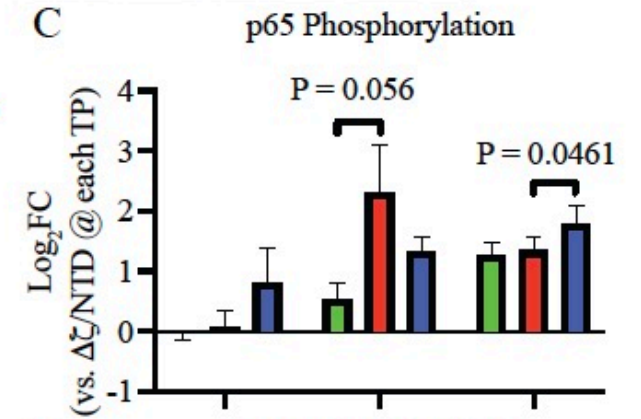
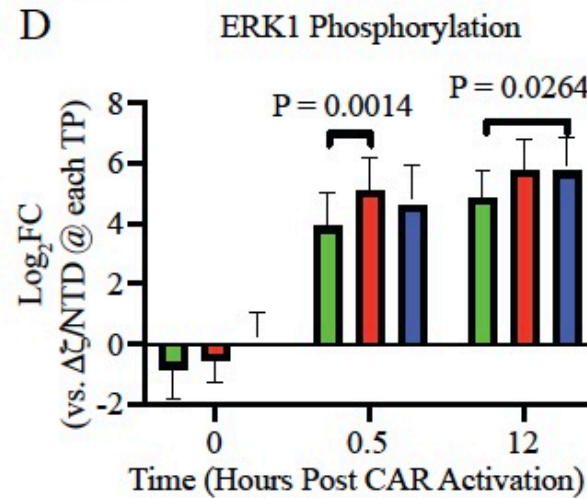
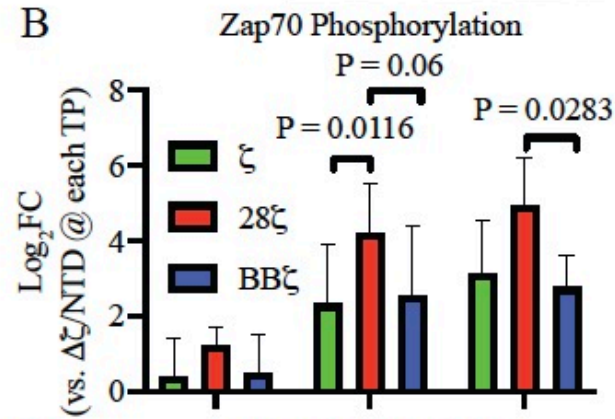
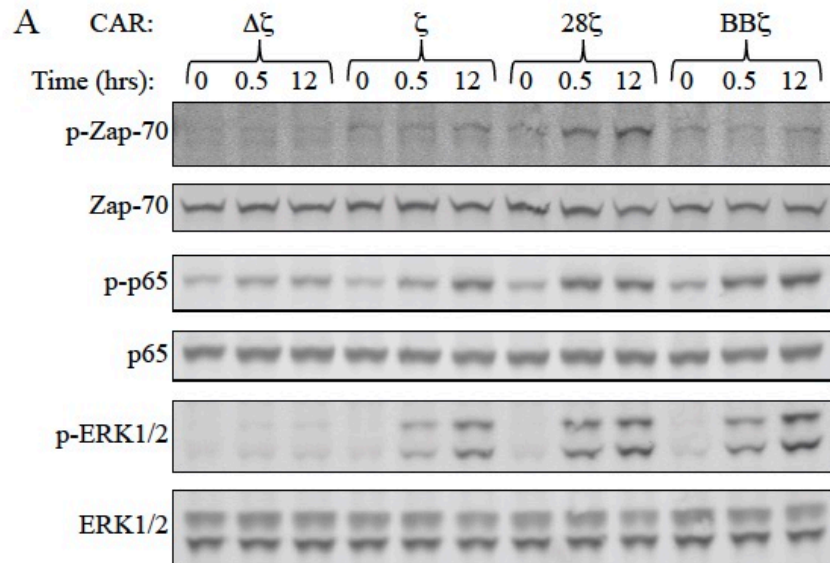
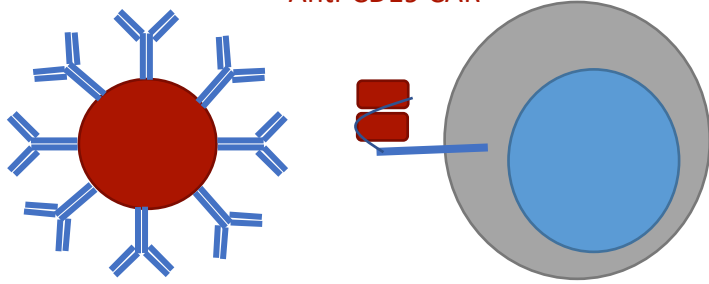
4-1BB-costimulated CARs induce greater clonal T cell expansion through increased cell survival



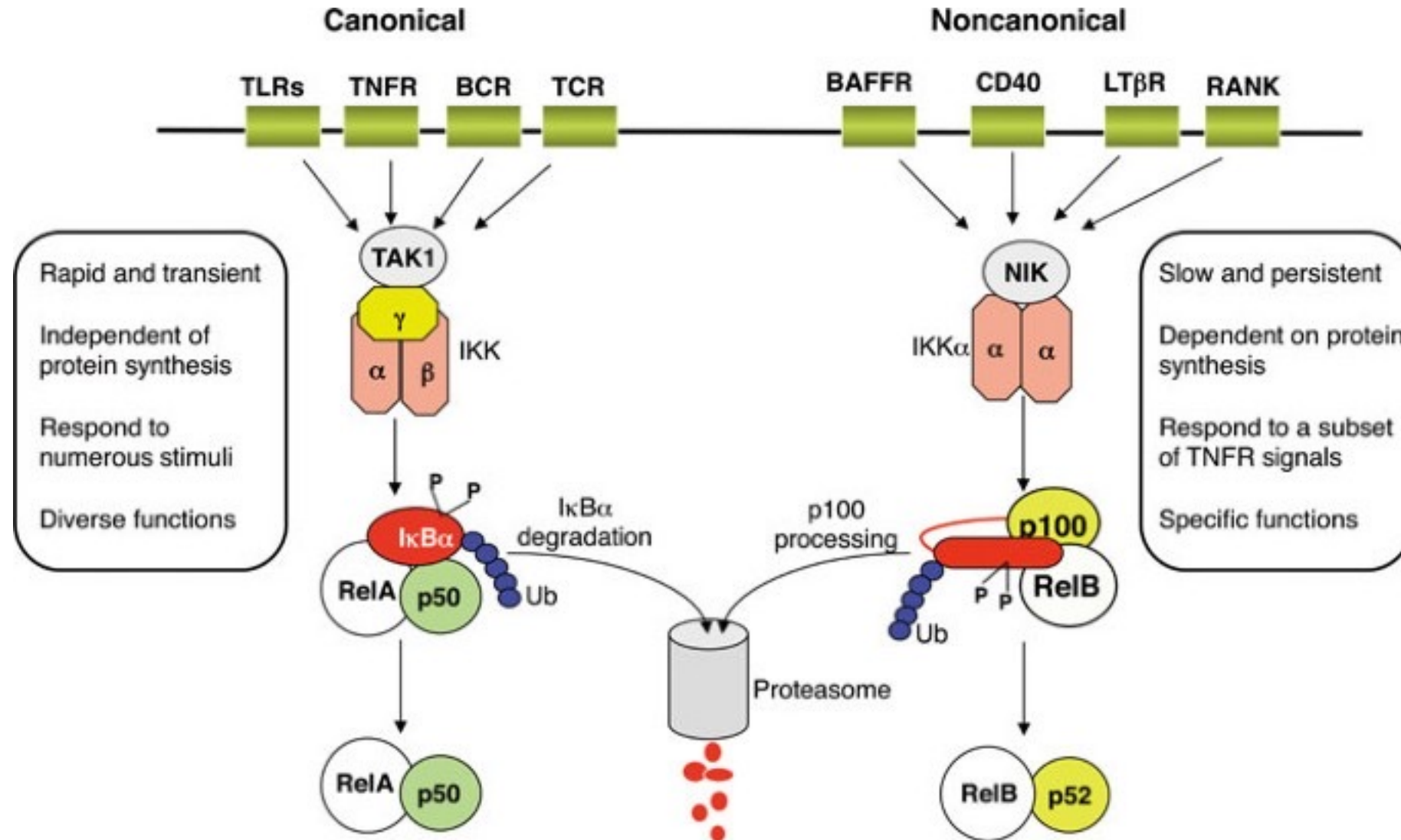
Establishing a bead-based CAR activation system for studying CAR signaling

anti-FMC63 idiotype

Anti-CD19 CAR

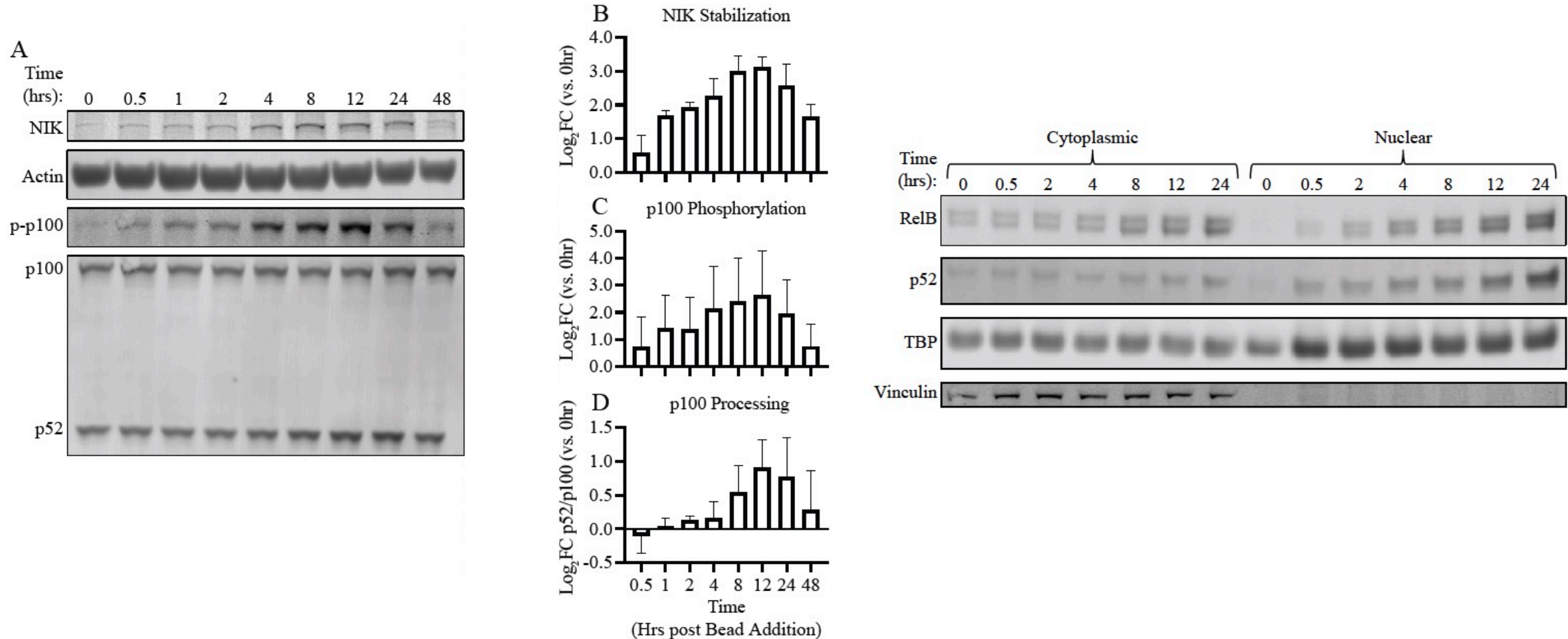


Canonical vs. Non-canonical (alternative) NF- κ B Pathways

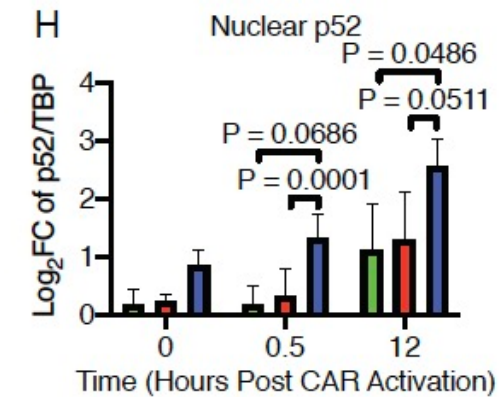
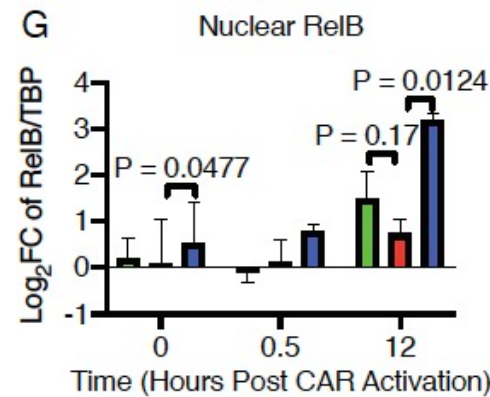
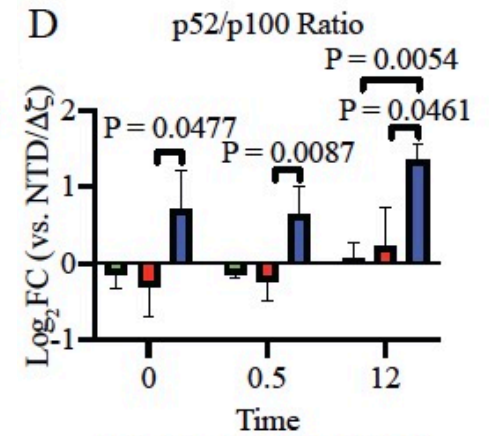
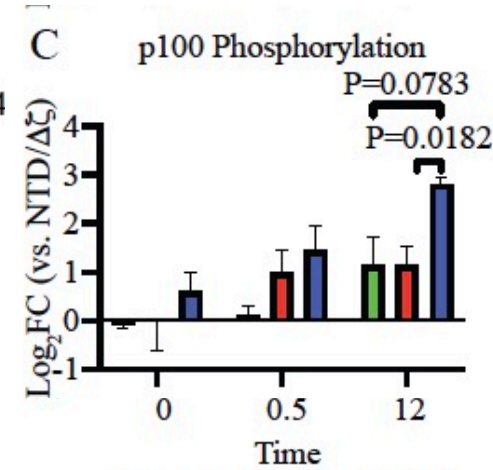
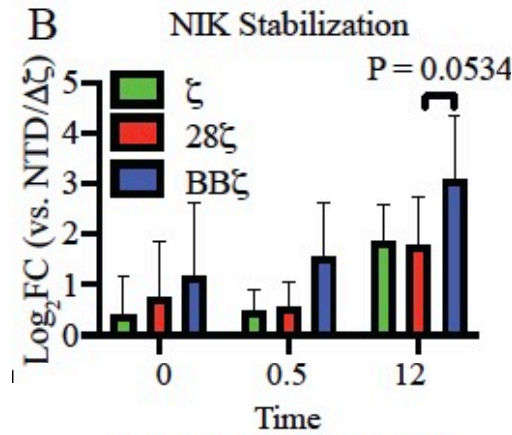
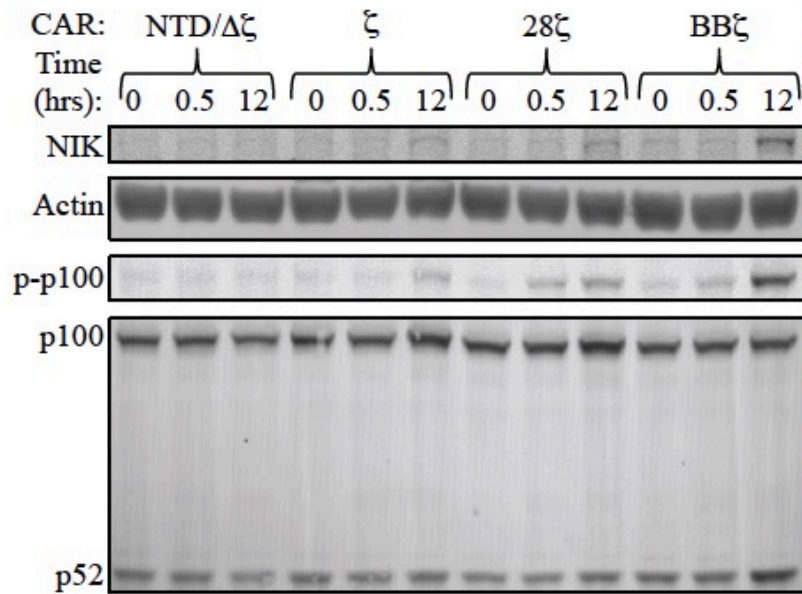


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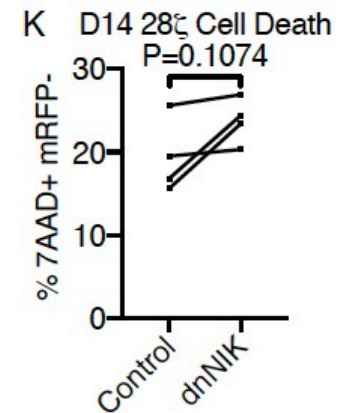
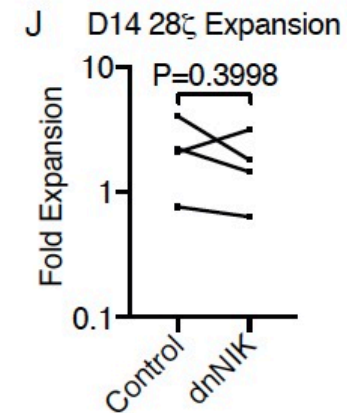
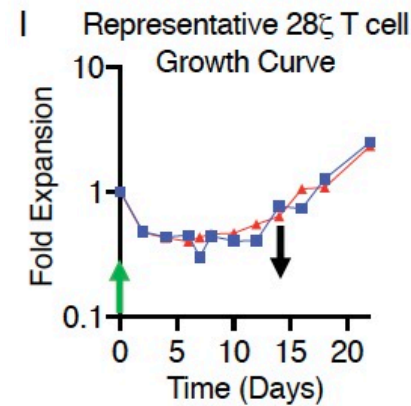
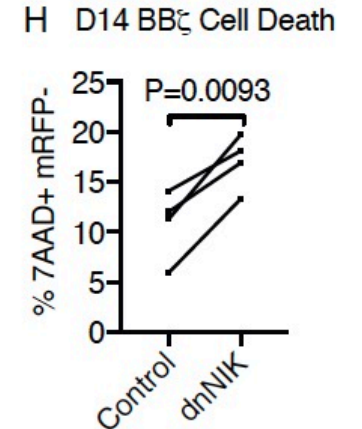
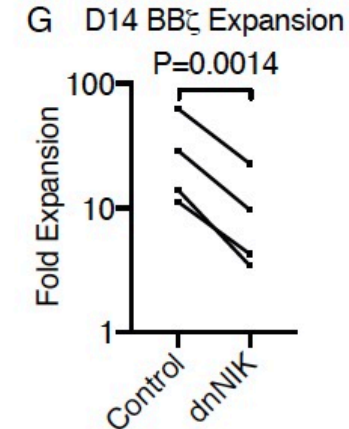
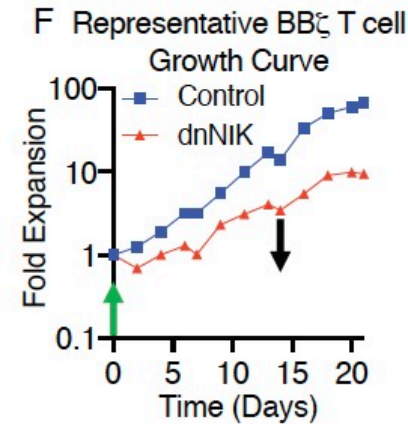
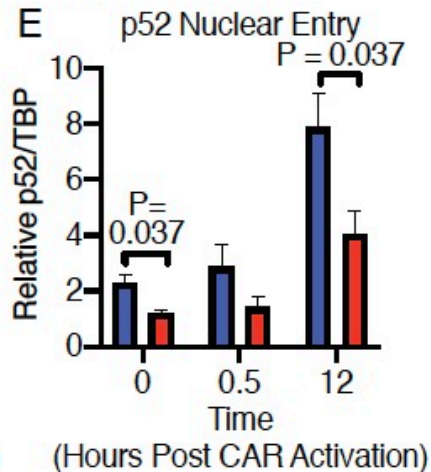
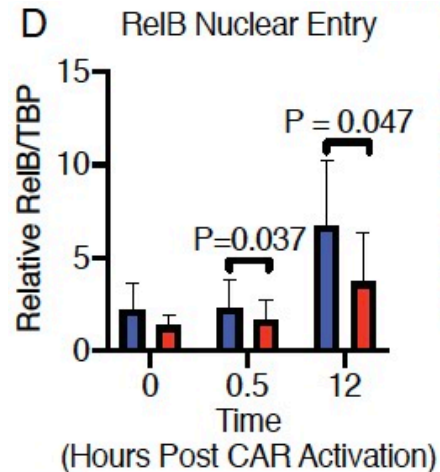
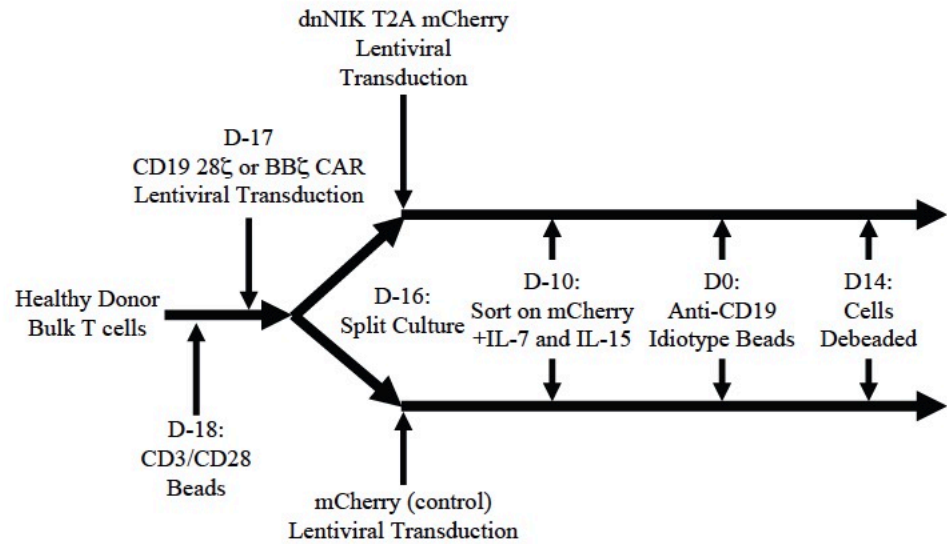
BB ζ CAR activates the non-canonical NF- κ B pathway



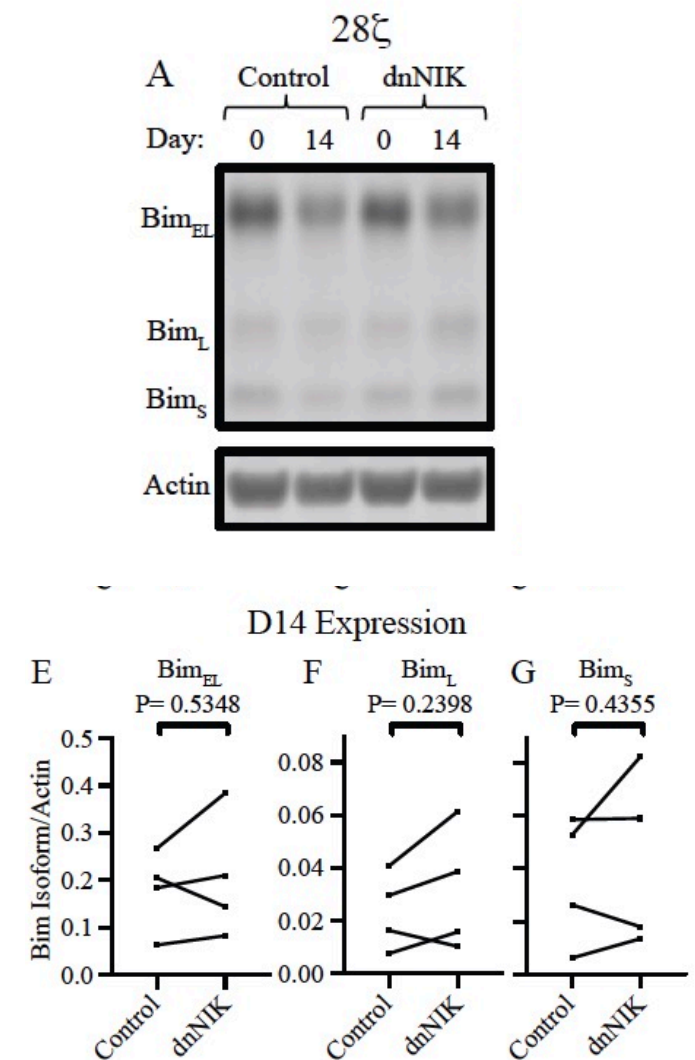
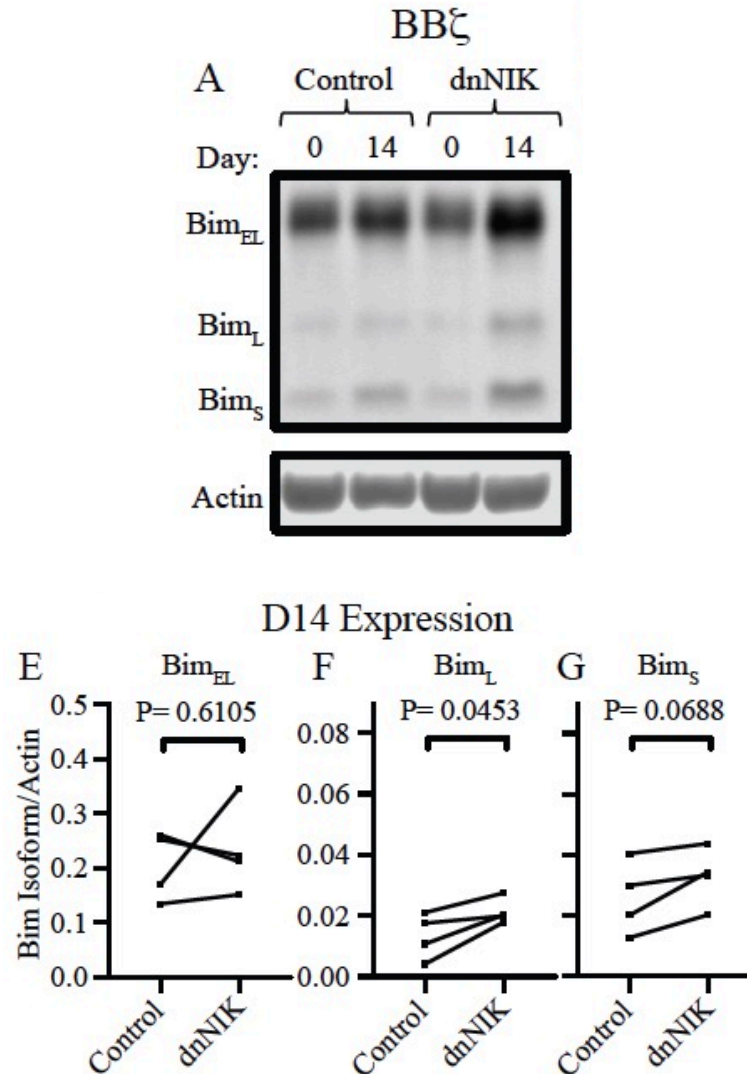
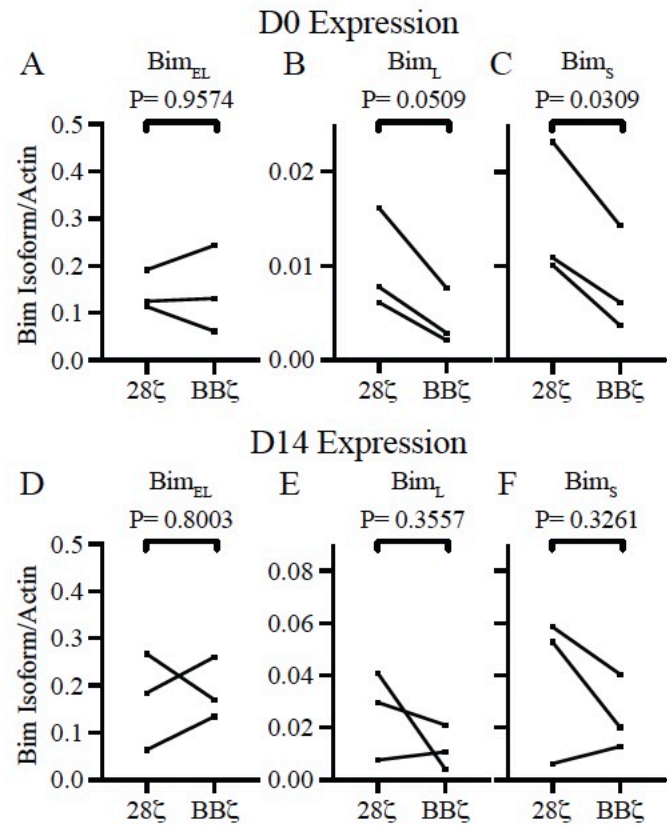
BB ζ CAR promotes greater $\text{ncNF-}\kappa\text{B}$ signaling compared with 28 ζ or ζ -only CARs



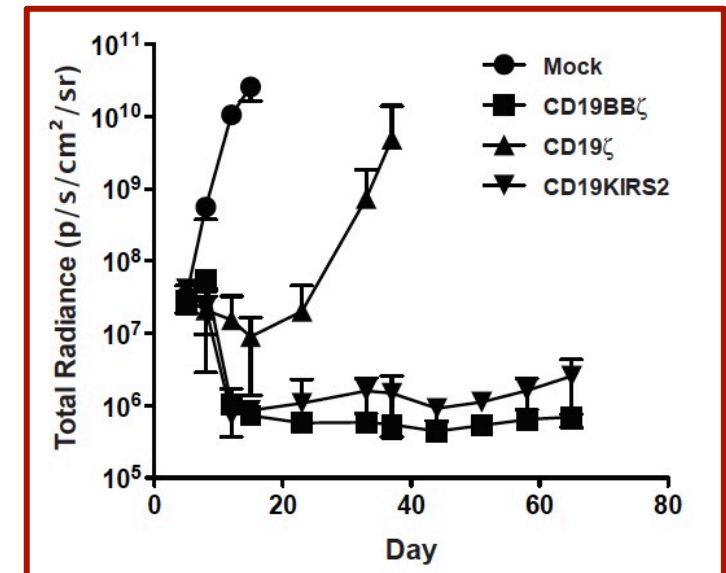
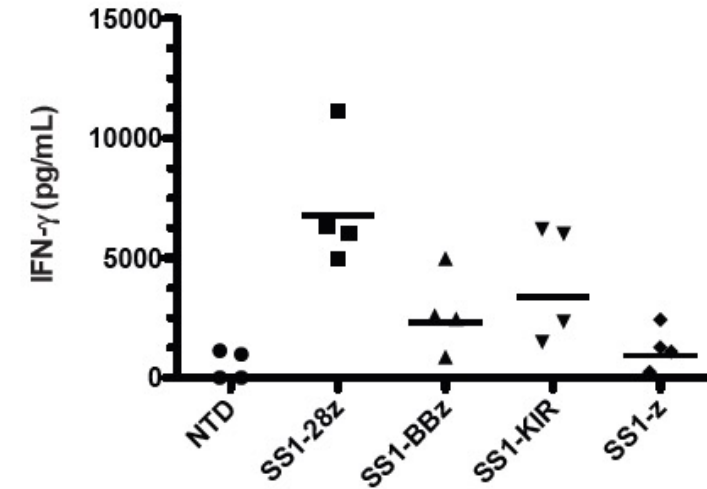
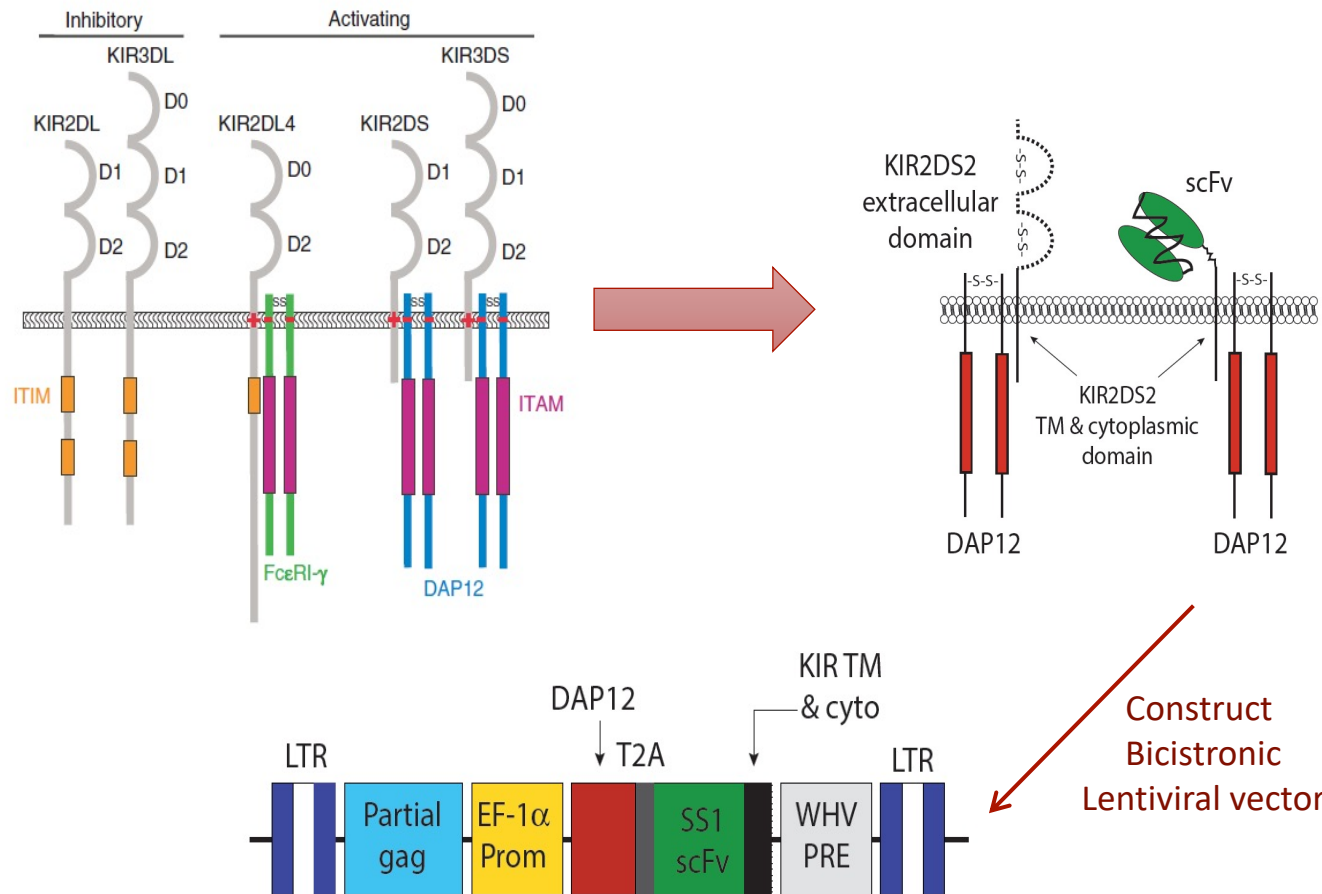
4-1BB-costimulated CAR T cells require ncNF- κ B signaling for survival and clonal expansion



4-1BB promotes T cell survival through suppression of proapoptotic Bim expression

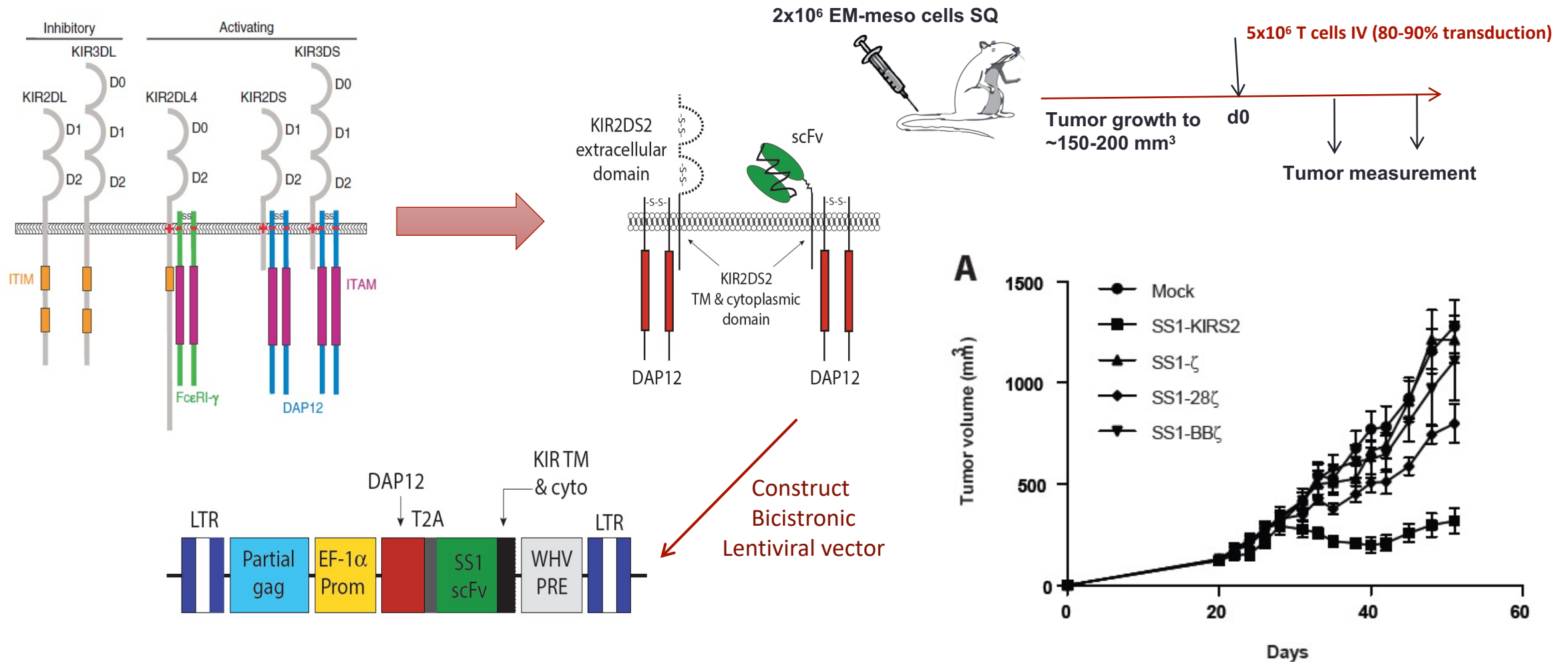


Is 4-1BB costimulation through a CAR ideal for all tumor settings?

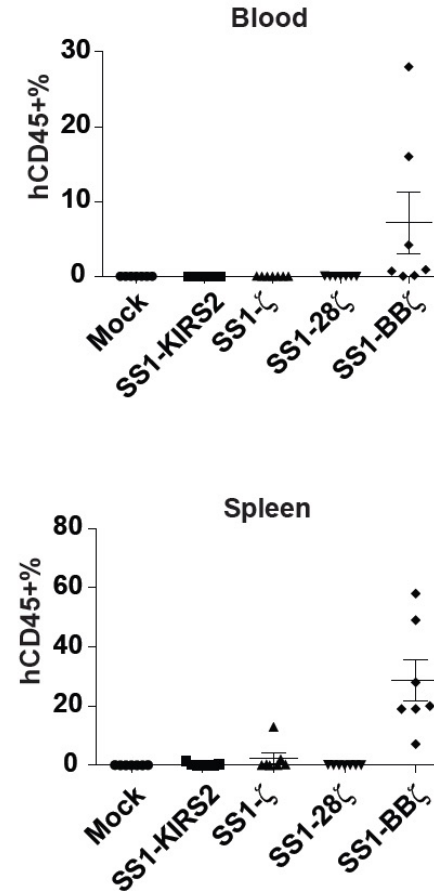
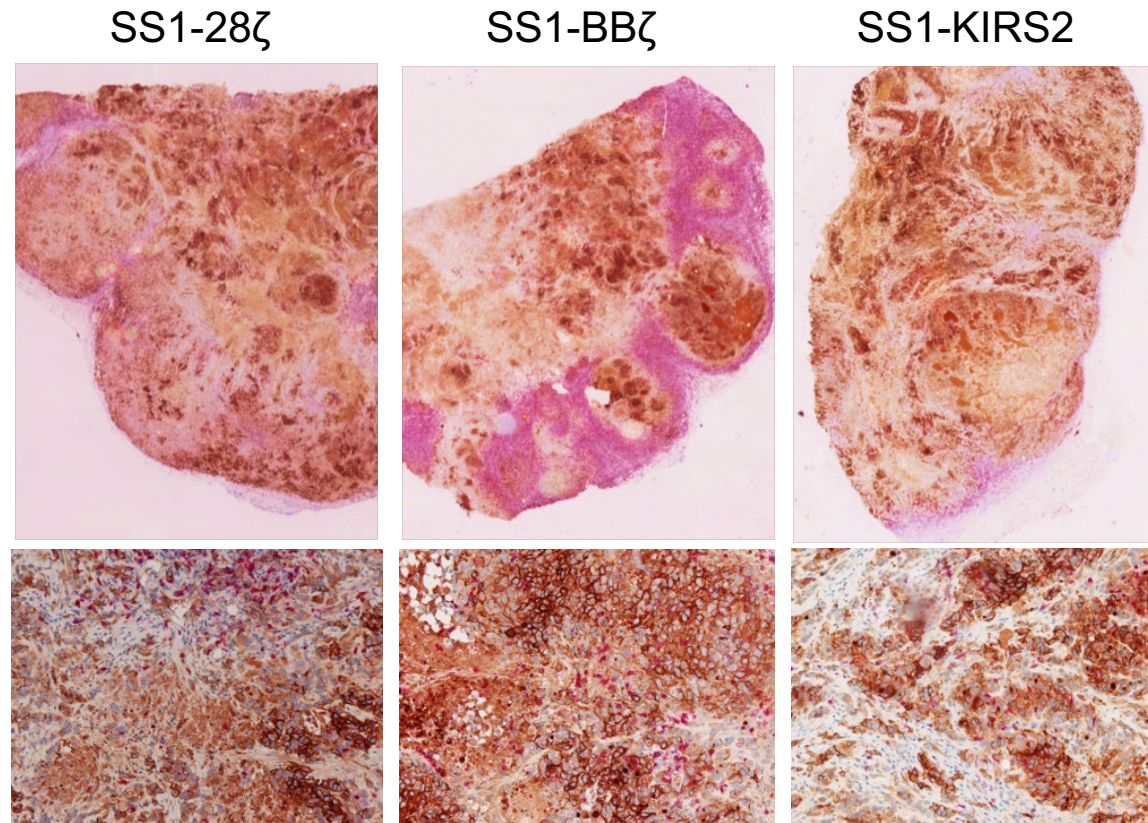


Wang et al. Cancer Immunol Res. 2015 3(7):815-26

Is 4-1BB costimulation through a CAR ideal for all tumor settings?

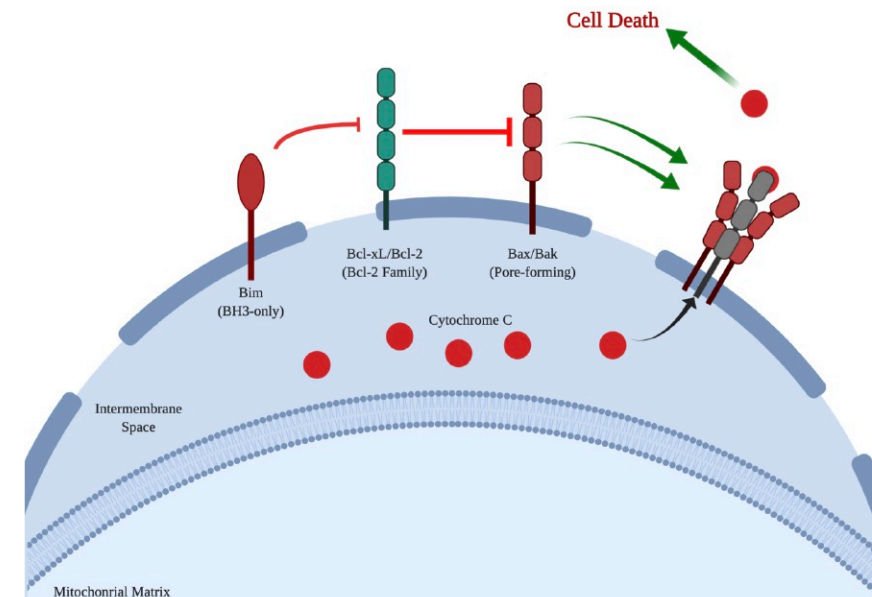
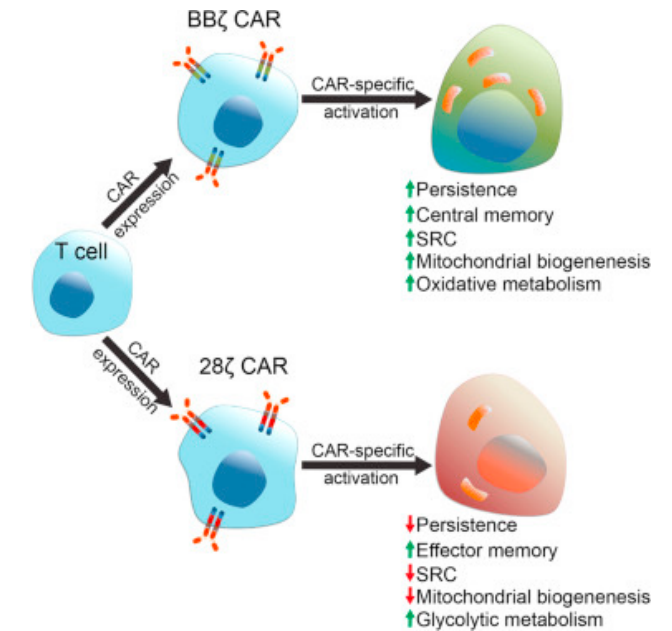


BB ζ costimulated CAR T cells show excellent persistence despite failing to control tumor



Summary

- CARs with a 4-1BB domain promote T cell persistence following adoptive transfer
 - Persistence is associated with enhanced anti-leukemic responses using a CD19-specific CAR.
 - Persistence does not lead to enhanced antitumor activity in some solid tumor models.
- 4-1BB promotes generation of T cells with mitochondrial mass and a more oxidative metabolism.
- 4-1BB signals in a CAR promote T cell survival through multiple signaling pathways including Erk and NF- κ B.
- ncNF- κ B is a unique pathway in 4-1BB-costimulated CAR T cells that promotes T cell survival through suppression of Bim expression, an important cellular pro-apoptotic gene.



Colleagues and Collaborators

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The Common Fund