

# Eradication of Established CD19+ Leukemia using a Single Injection of Chimeric Immunoreceptor Modified Lentiviral Transduced T Cells in a Xenograft NOG Mouse Model

David M. Barrett\*, Carmine Carpenito\*, Yang Bing Zhao,  
Michael Kalos, Stephan A. Grupp and Carl June.

Division of Oncology, Children's Hospital of Philadelphia,  
Philadelphia, PA and the Abramson Family Cancer  
Research Institute, University of Pennsylvania,  
Philadelphia, PA



# Disclosure

- The author has no relevant financial disclosures or conflicts of interest.
- This involves no off label use of any medications.

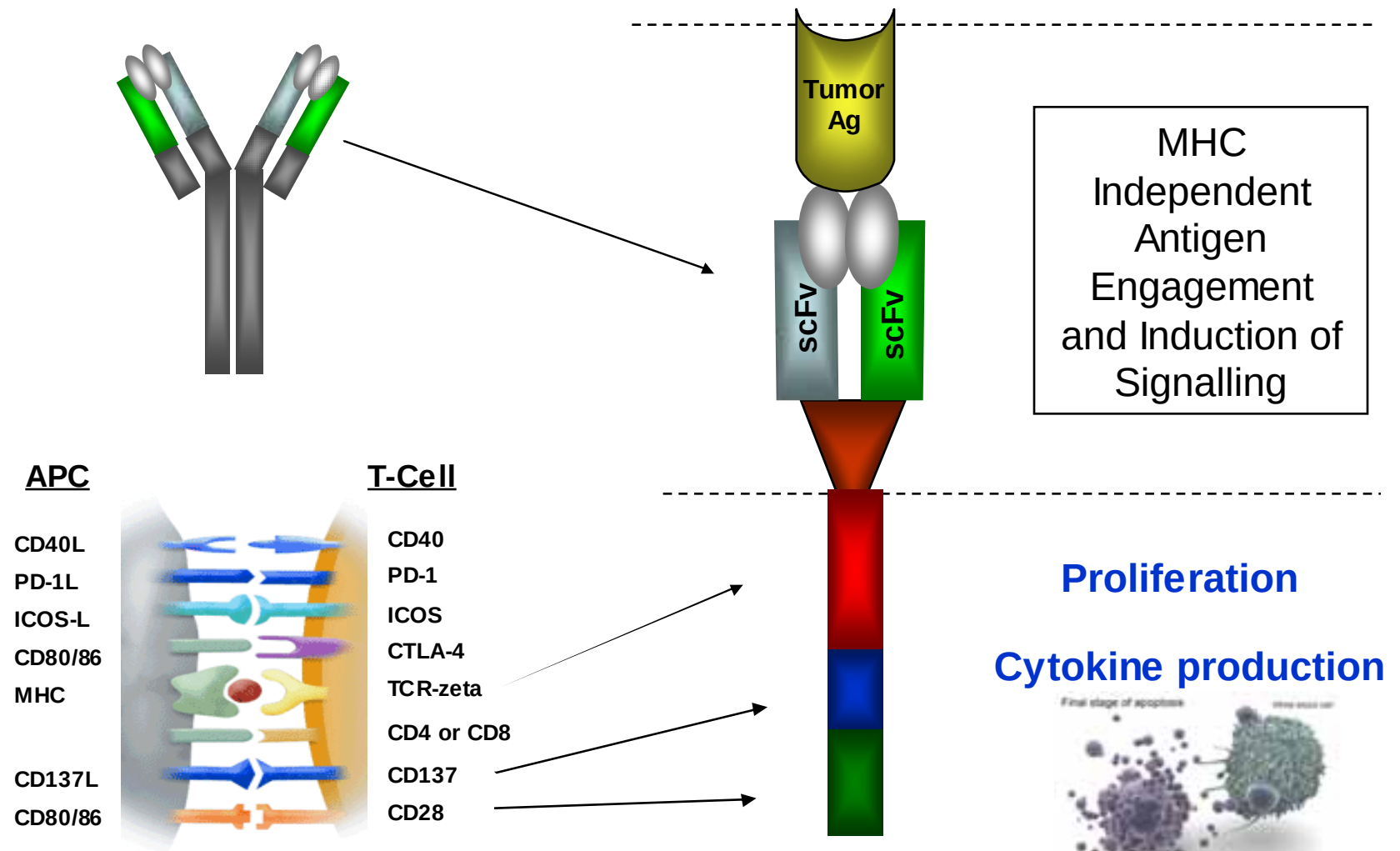


# Overview

- Acute Lymphoblastic Leukemia (ALL) is the most common childhood malignancy, and relapsed ALL is the fifth most common as well as the most common cause of cancer death in children
- Higher doses of chemotherapy for relapsed/refractory disease come with increased toxicity (and not that much more success)
- Donor derived T-cells studied as part of the Graft Versus Leukemia Effect in allogeneic transplant are effective in eradicating hematologic malignancies but at the cost of Graft Versus Host disease
- Chimeric Immunoreceptors are artificial constructs fusing the extracellular antigen-recognition portion of an immunoglobulin with activation and costimulatory domains of T-cell receptors
- Chimeric Immunoreceptors expressed in Autologous T-cells provide the potential for cytotoxic efficacy without Graft Versus Host disease



# Chimeric Immunoreceptor



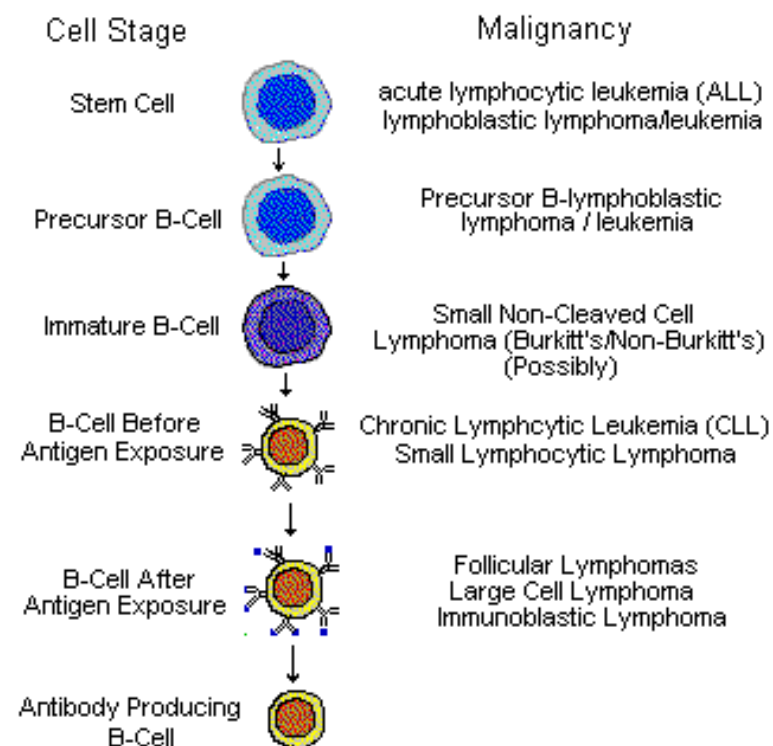
Carreno and Collins. *Annu Rev Immunol.* 2002;20:29.  
Stuart and Racke. *Expert Opin Ther Targets.* 2002;6:275.



# CD19 as a tumor target antigen

- CD19 is expressed by the majority of B-cell derived tumors
- Normal CD19 expression is limited to only B-lineage lymphoid cells
- B-cell deficiency is well tolerated given extensive experience with rituximab (anti CD20 antibody), and is treatable with IV immunoglobulin

## B Cell Cancers by Cell Development



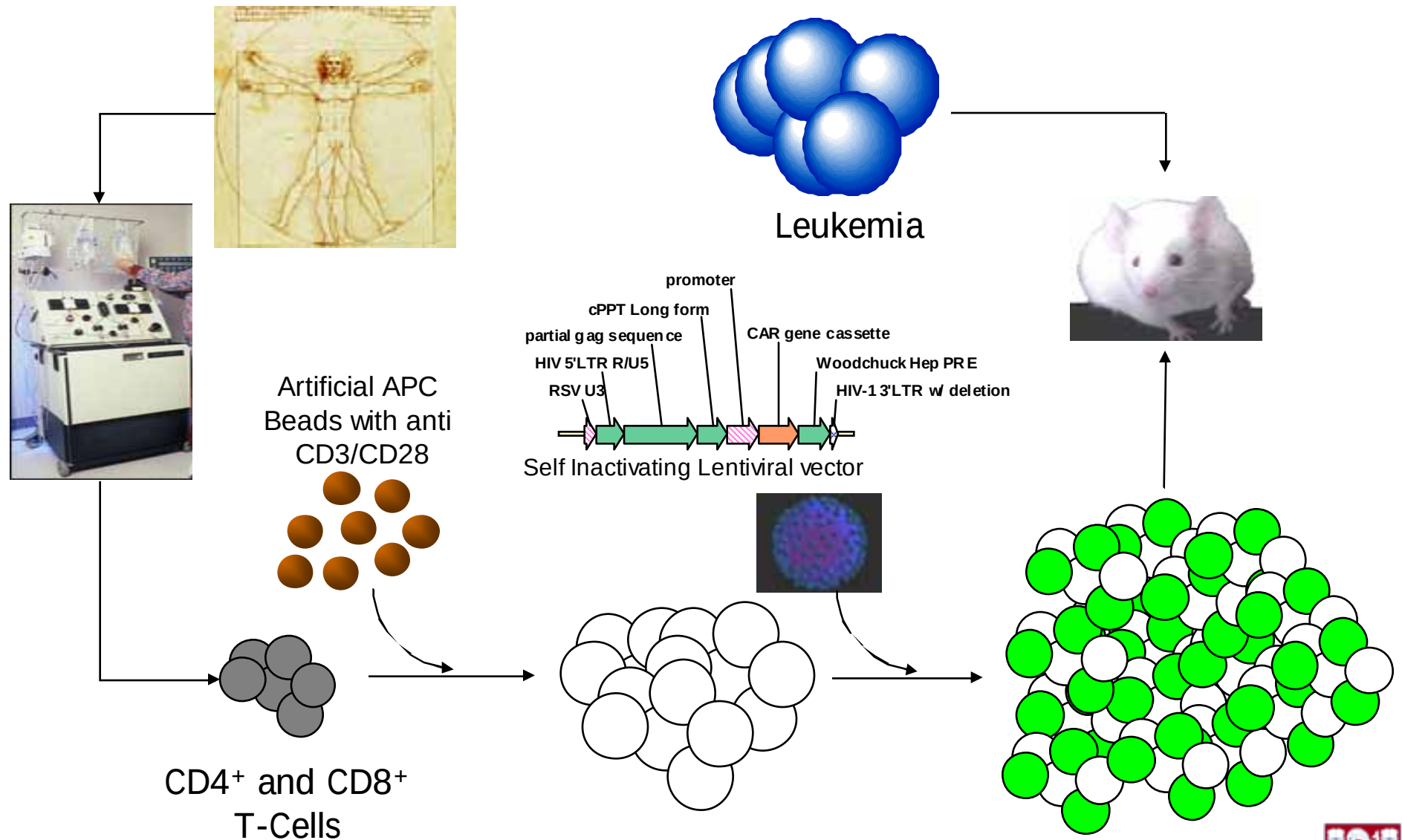
Lymphoma Information Network  
<http://www.lymphomainfo.net/>

# Incorporating co-stimulatory signals

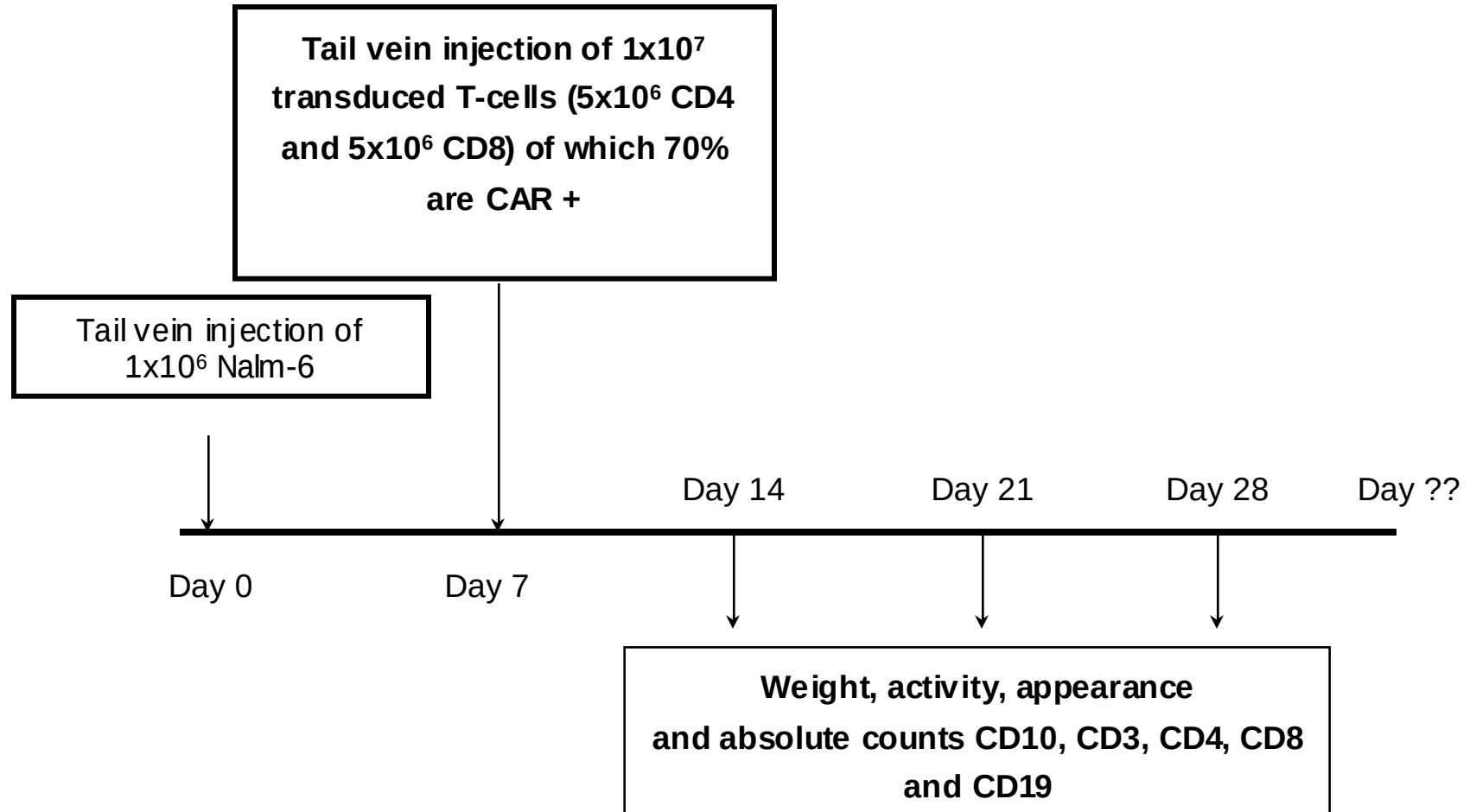
- CD28 provides potent activating signals that enhance CD4 and CD8 T cell:
  - Proliferation
  - Survival
  - Cytokine production
- 4-1BB (CD137) provides signals that:
  - Promote long-term proliferation and survival of CD8 T cells
  - Promote cytokine production
  - enhance CD8+ T cell responses in viral infection and allograft rejection



# Pre-clinical Experimental Model

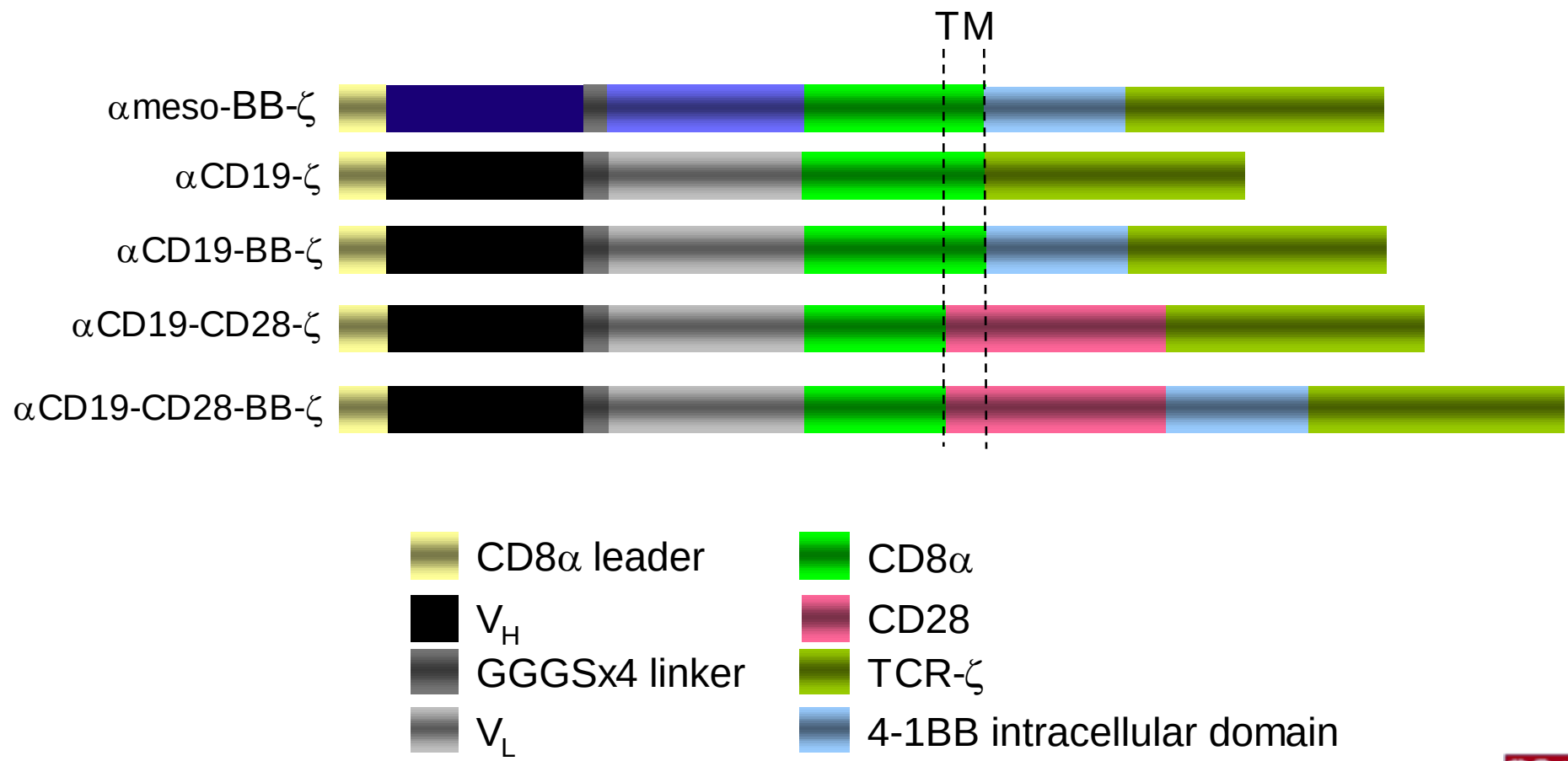


# Methods

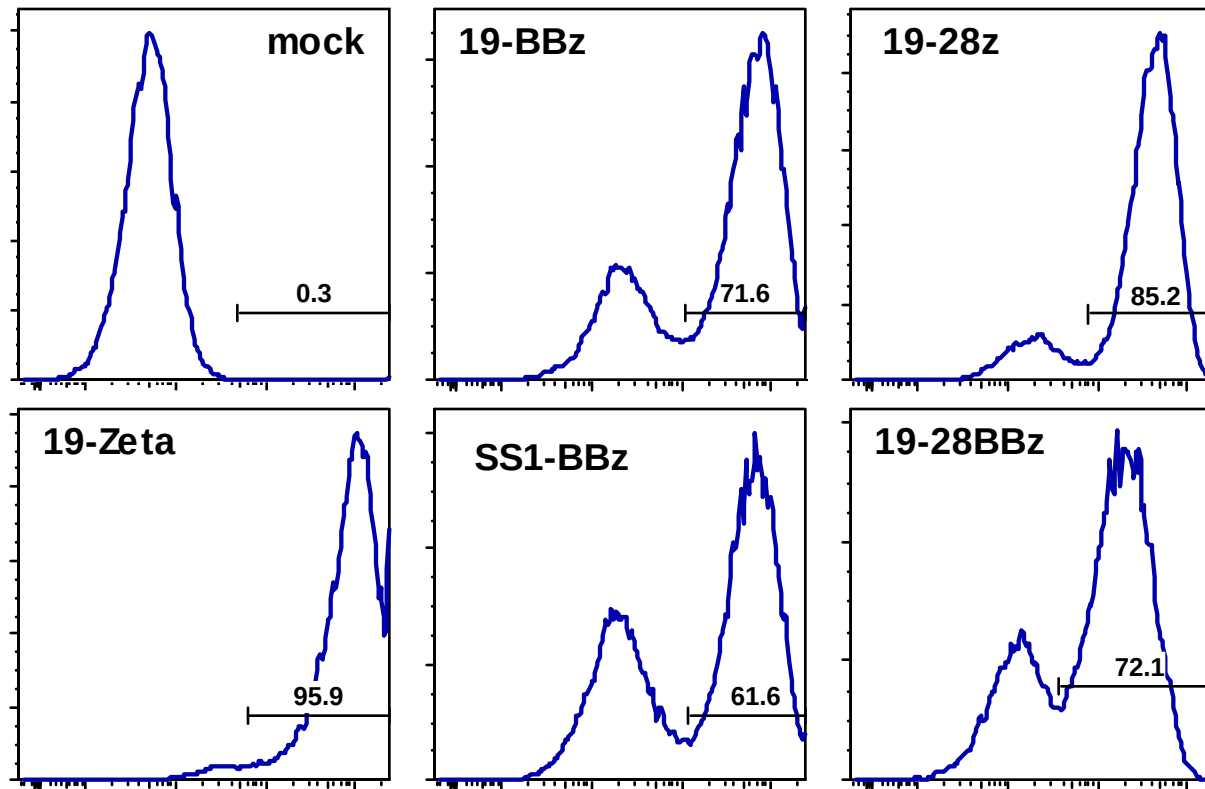




# Chimeric Immunoreceptors



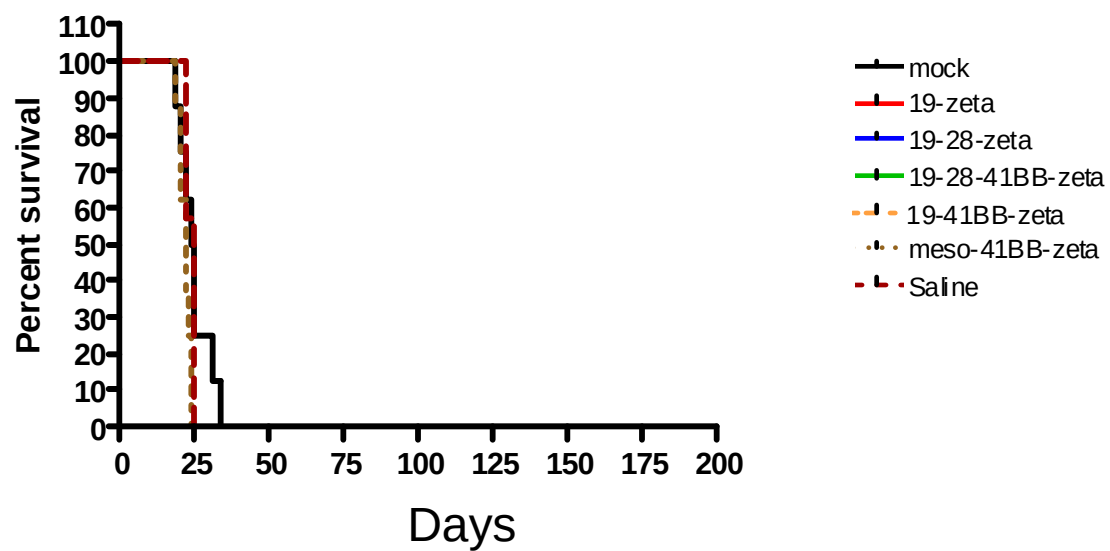
## ND150 CAR Positive T cells Day of Injection



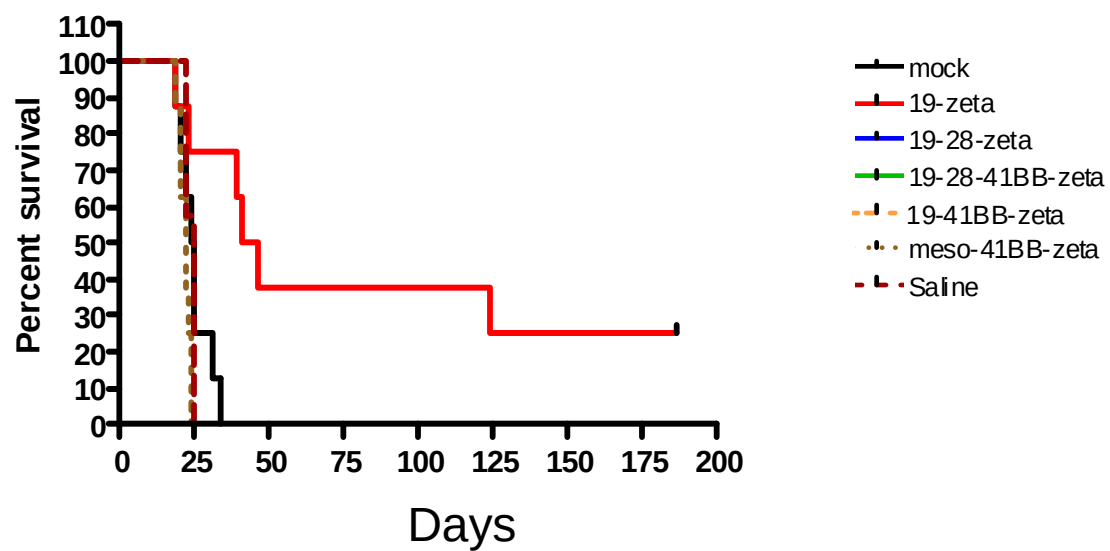
scFv expression



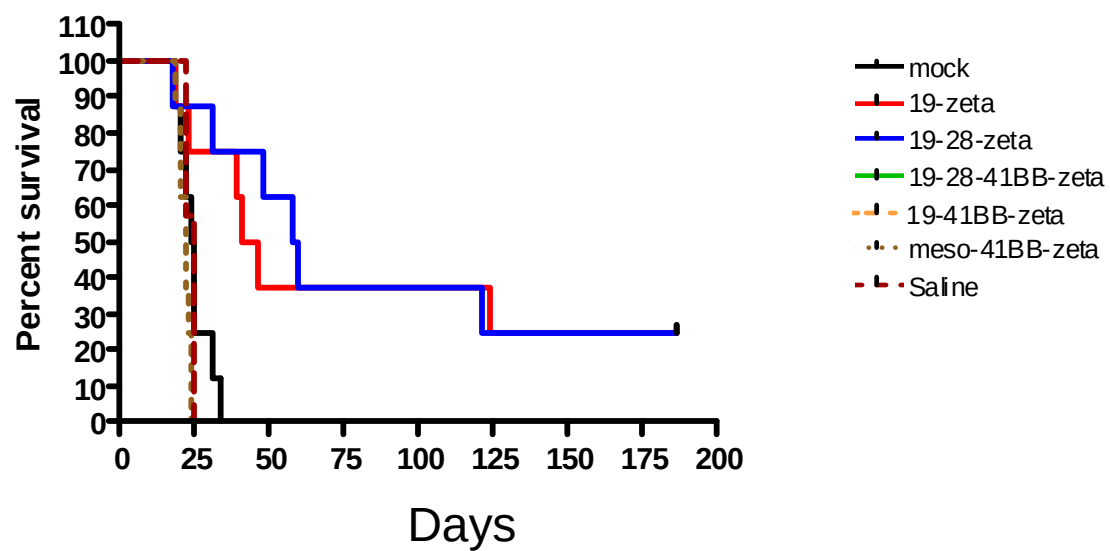
# Overall Survival



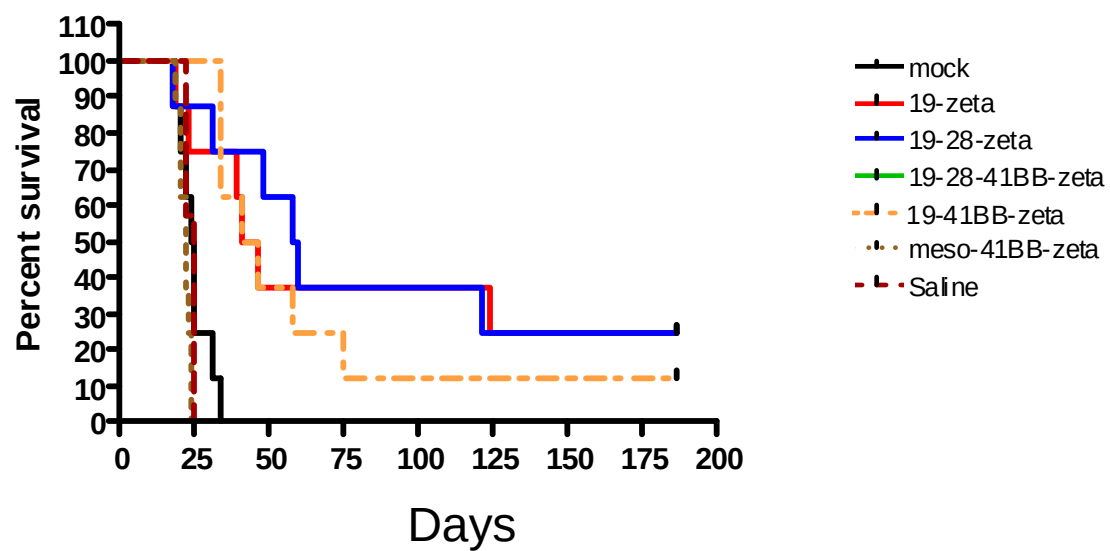
# Overall Survival



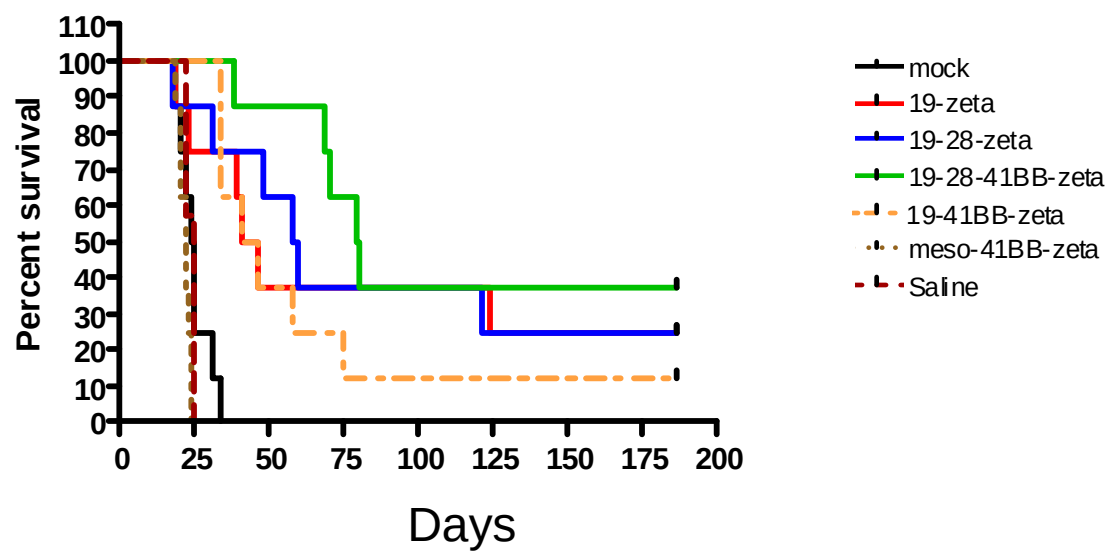
# Overall Survival



# Overall Survival

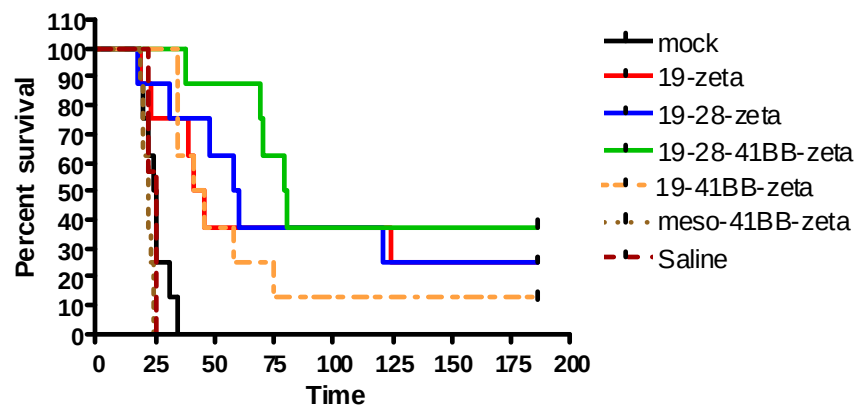


# Overall Survival

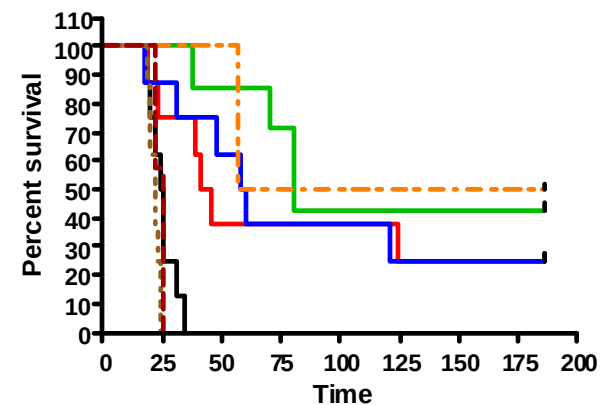


# Influence of Xenogeneic Graft Versus Host Disease

Overall Survival



Overall Survival –  
GVHD Excluded



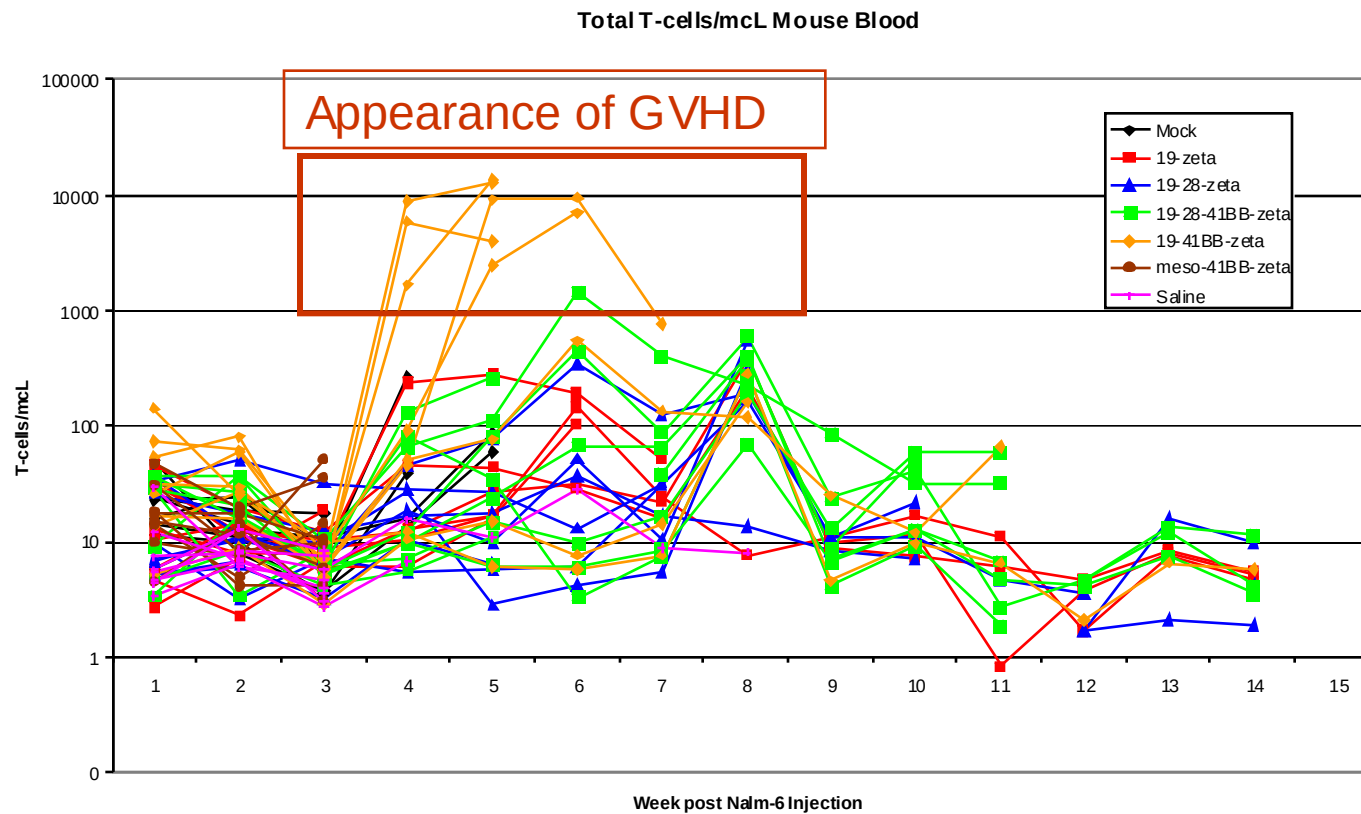
| Group           | Median OS | Median OS<br>(GVHD excluded) |
|-----------------|-----------|------------------------------|
| Saline          | 25        | 25                           |
| Mock            | 24        | 24                           |
| Meso-41BB-zeta  | 22        | 22                           |
| 19-zeta         | 41*       | 41*                          |
| 19-28-zeta      | 58*       | 58*                          |
| 19-41BB-zeta    | 41*       | 57                           |
| 19-28-41BB-zeta | 79*       | 80*                          |

\*p<0.001  
(mock)

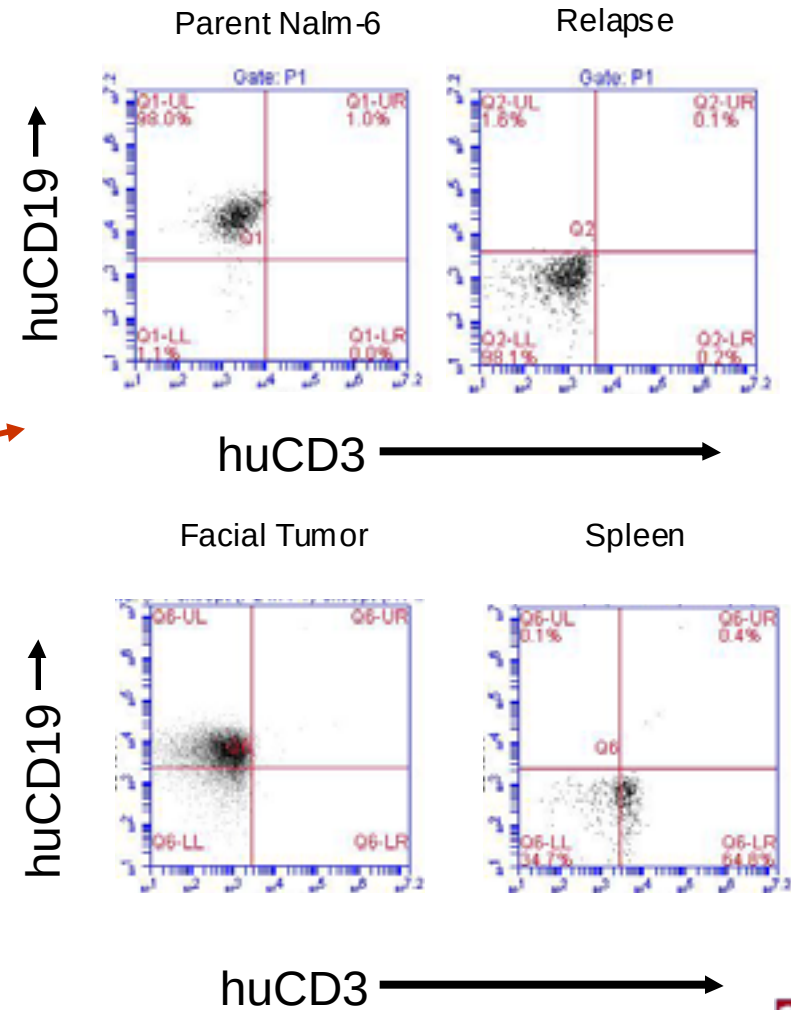
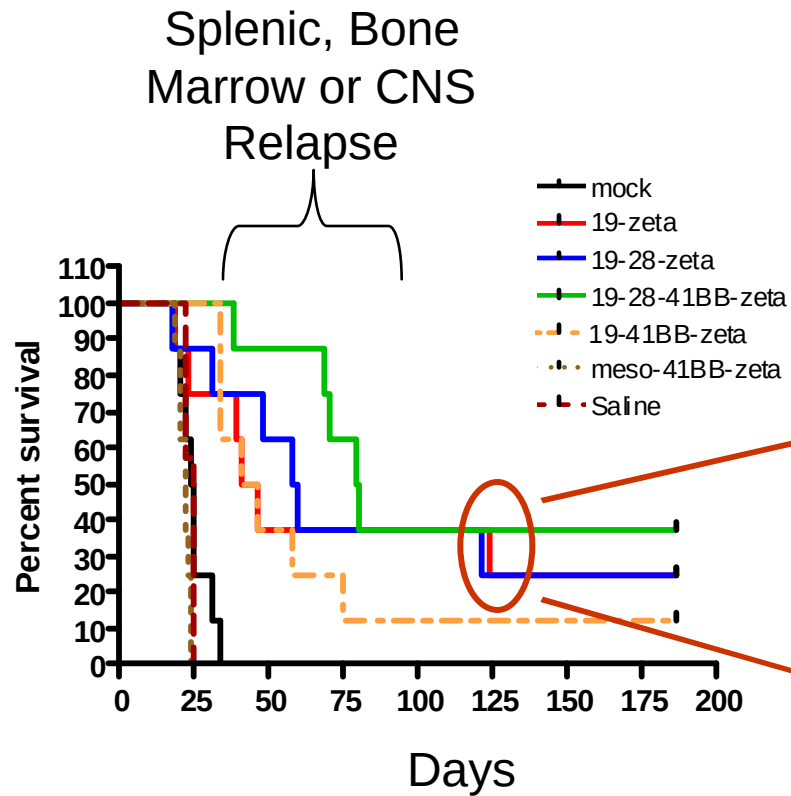




# Total Peripheral Blood T Cells correlate with onset of GVHD



# Disease Progression or Relapse



# Summary

- Immunoreceptor modified T cells generated by lentiviral transfection of *ex vivo* expanded T cells can effectively eradicate human leukemia in a xenograft NOG mouse model with only a single injection of  $1 \times 10^7$  T cells.
- Slight survival advantage over other constructs is observed with the 19-28-41BB- $\zeta$  CAR
  - Including apparent eradication of disease (survival >100 days post disappearance of circulating T cells, >60 days since any relapse in any other group)
- High total circulating T cells correlates with xenogeneic GVHD mortality in the absence of detectable leukemia



# Acknowledgements

## Grupp Lab

- Stephan Grupp
- Junior Hall
- Terri Ryan
- Alix Seif
- Gregor Reid
- Cecelia Sheen
- Jessi Hulitt
- Eliza Strong

## June Lab

- Carl June
- Carmine Carpenito
- Yang Bing Zhao
- Michael Kalos

