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"Advancing CAR T Cell Therapy for the Treatment of Recurrent Ovarian Cancer"

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Assistant Professor, Hematology/HCT and Immuno-Oncology

Beckman Research Institute

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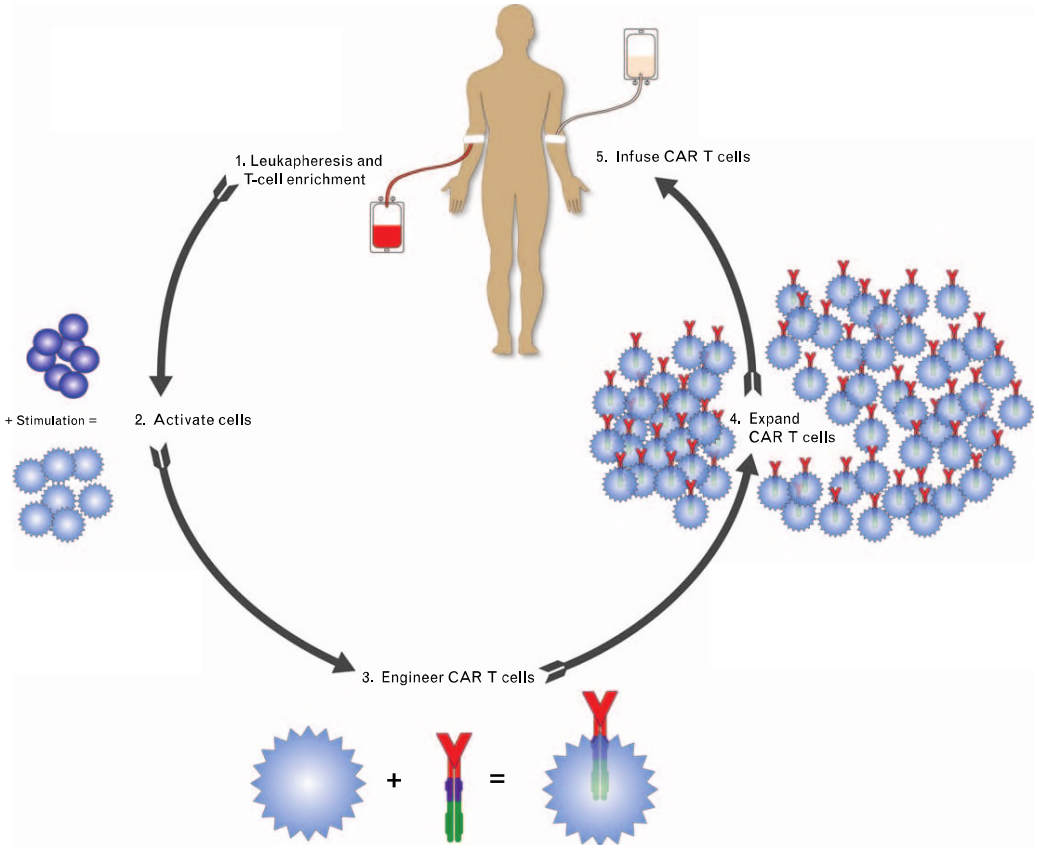
CAR T Cell Therapy for Solid Tumors



Prostate NCT03873805

Prostate Cancer Foundation
Curing Together.

Breast-Brain Metastasis NCT03696030

CIRM
CALIFORNIA'S STEM CELL AGENCY

Ovarian NCT05225363

NIH R01

Priceman et al. *Curr Opin Immunol* 2015

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Challenges Facing CAR T Cell Therapies for Solid Tumors

- Tumor antigen heterogeneity
- Immunosuppressive tumor microenvironment (TME)

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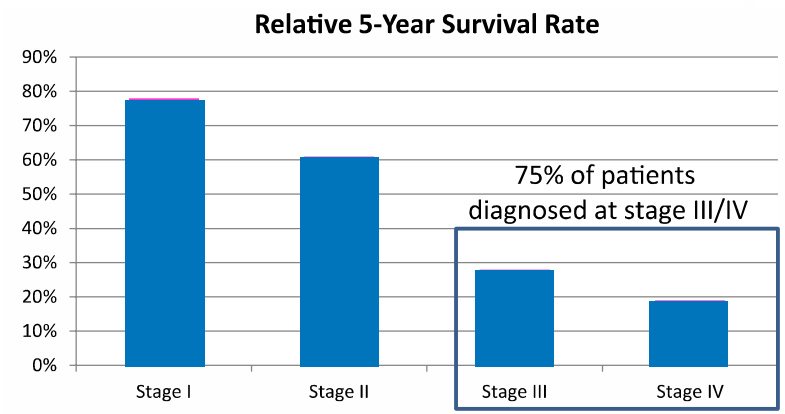
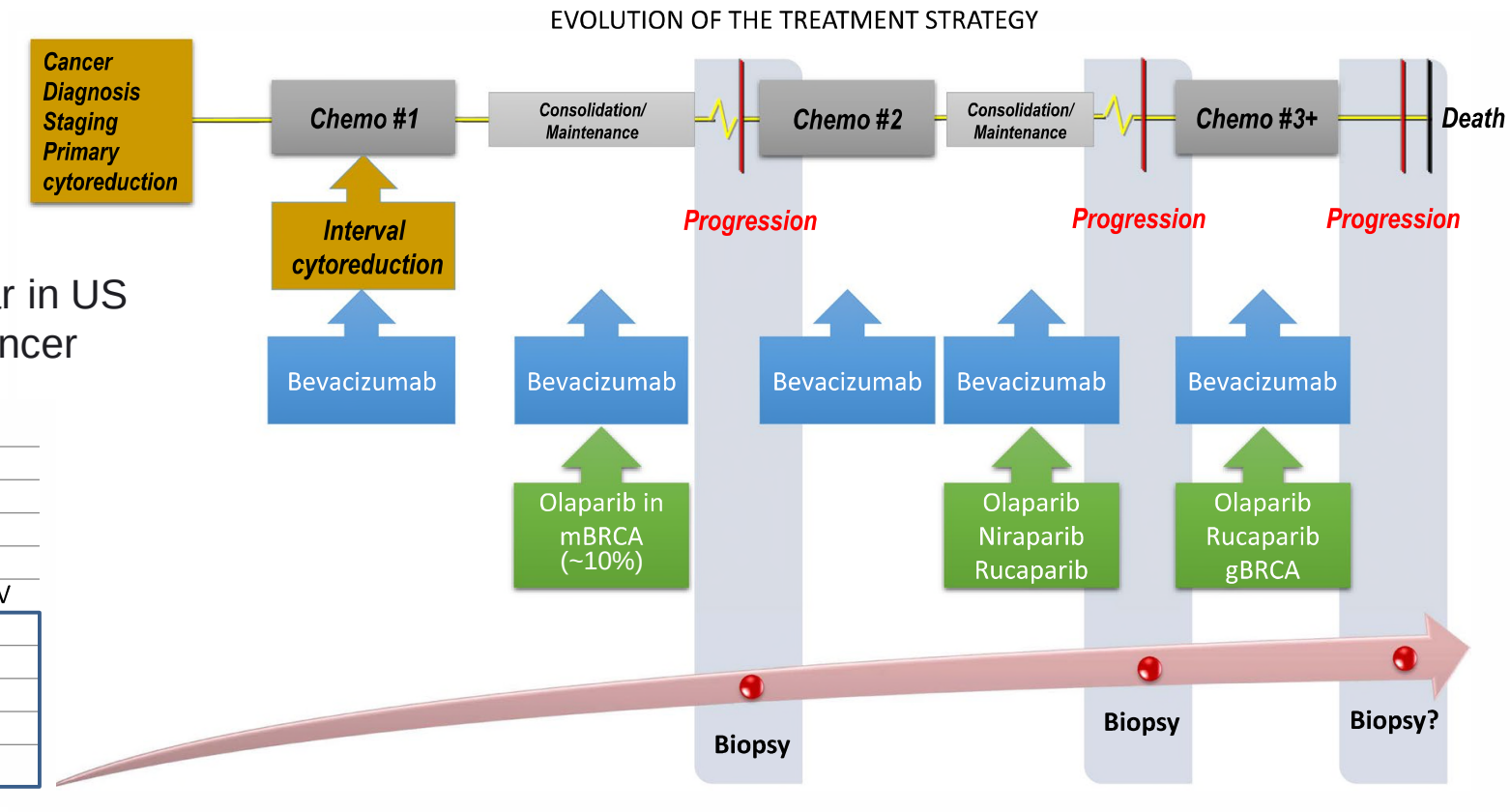
Ovarian Cancer Incidence and Treatment Landscape

~19,880 women new diagnoses per year in US
~12,810 women will die from ovarian cancer





Adapted from Lheureux et al., *CA Cancer J Clin,* 2019

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Ovarian Cancer CAR Targets: Aberrant Glycosylation

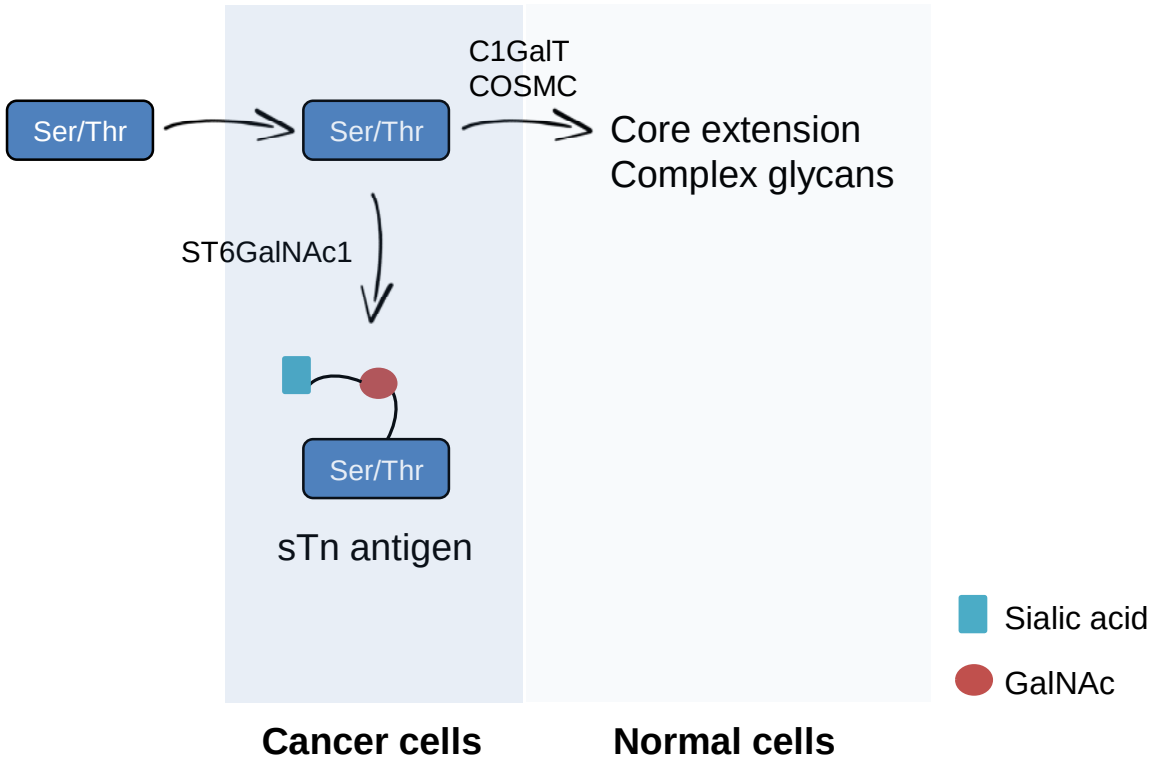
Hypothesis: Post-translational modification targets may change antigen escape kinetics compared with transcriptionally-regulated targets, and therefore impact CAR T cell durable therapeutic responses

Adapted from Reily et al., *Nature Reviews,* 2019

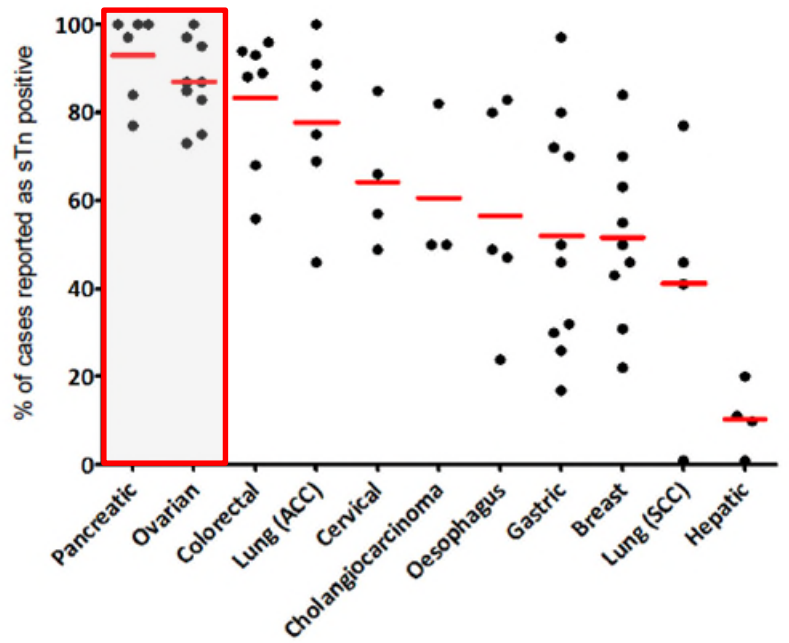
Tumor-Associated Glycoprotein-72 (TAG72) as a CAR target

Cancer Specific Glycosylation:

- Tn - O-glycans
- **STn – truncated O-glycans (e.g., TAG72)**



Reported TAG72 expression:



Adapted from Arabi et al., *Exp. Cell Research,* 2018

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Experience with TAG72 as a Therapeutic Target

- TAG72 is a pan-cancer antigen expressed in several tumor types including **ovarian**, breast, gastric, colorectal, endometrium, esophagus, and pancreas

- Theranostics target for multiple cancer types

- A first generation TAG72-CAR construct was made by Cell Genesys Inc over 10+ years ago. A phase 1/2 trial was conducted for stage IV colorectal cancer with liver metastases.
  - Safety with no DLTs at doses of 10^{10} TAG72-CAR T cells
  - Reduction in TAG72 serum markers
  - Anti-tumor activity in one patient exhibiting stable disease, but no responses in advanced patients (Hege, et al JITC 2017)

- **Can a next-generation TAG72-CAR T cell offer therapeutic benefit for patients with ovarian cancer peritoneal metastasis?**

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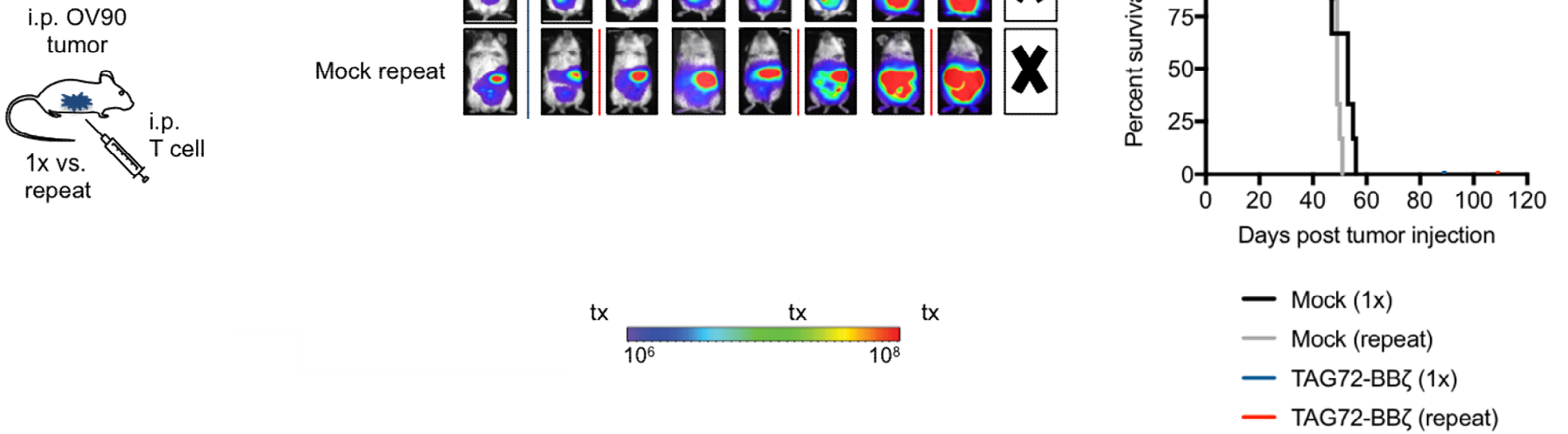
TAG72-CAR T Cells Target Ovarian Cancer Peritoneal Metastasis

John Murad
PhD



-**Repeat intraperitoneal** delivery of TAG72-CAR T cells extends controls of peritoneal ovarian tumors

Murad, et al. *Front Immunol* 2018

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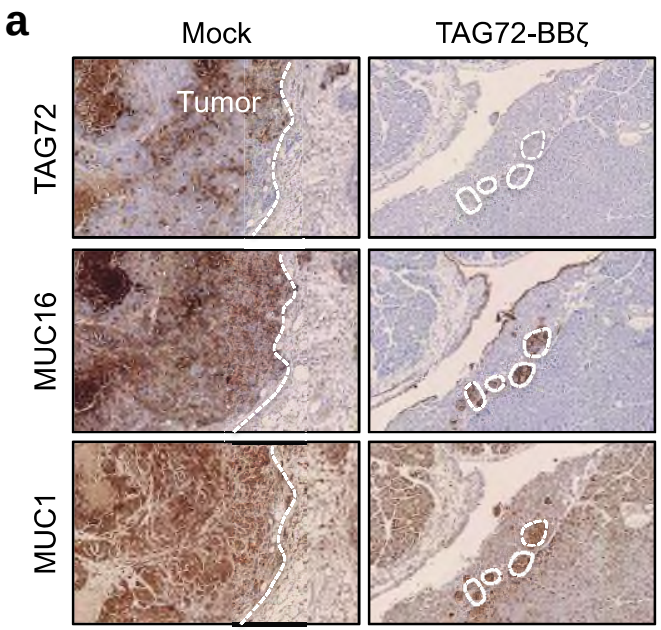
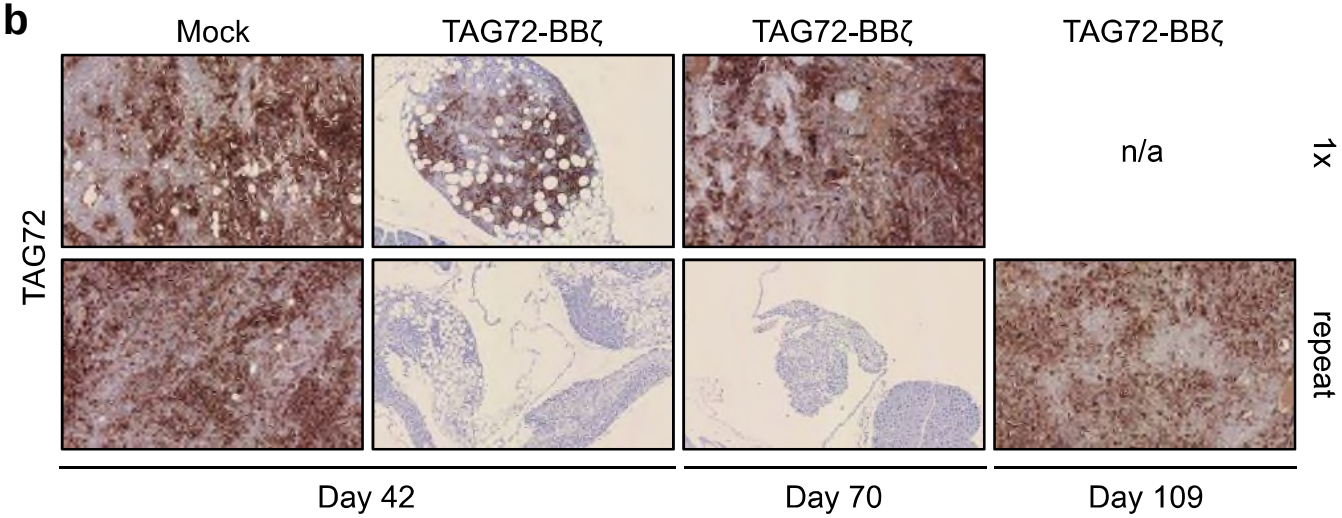
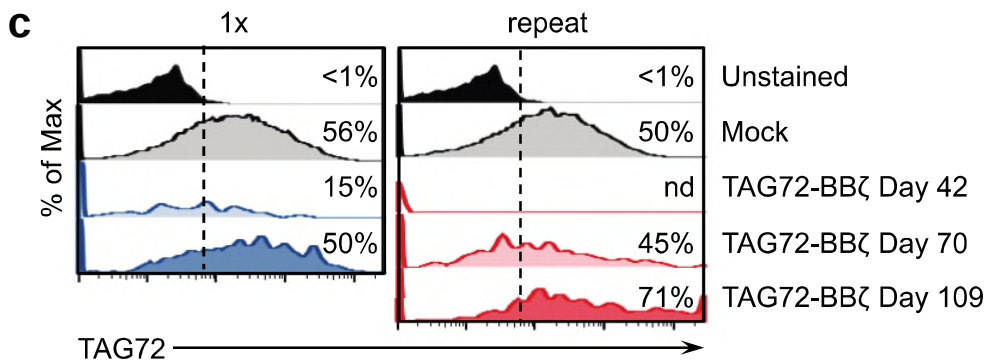
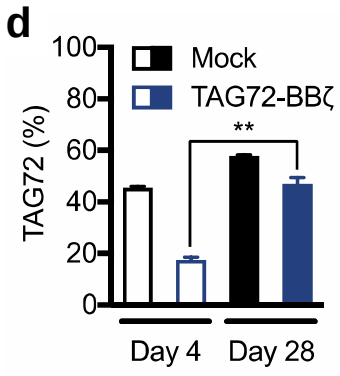
Is Antigen Escape Responsible for Tumor Resistance?





-TAG72 expression is transiently downregulated in early recurrences post CAR T cell therapy, but re-emerges in late recurrences





Murad, et al. *Front Immunol* 2018

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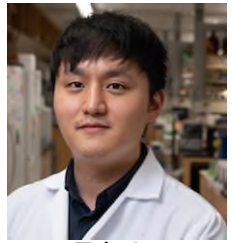
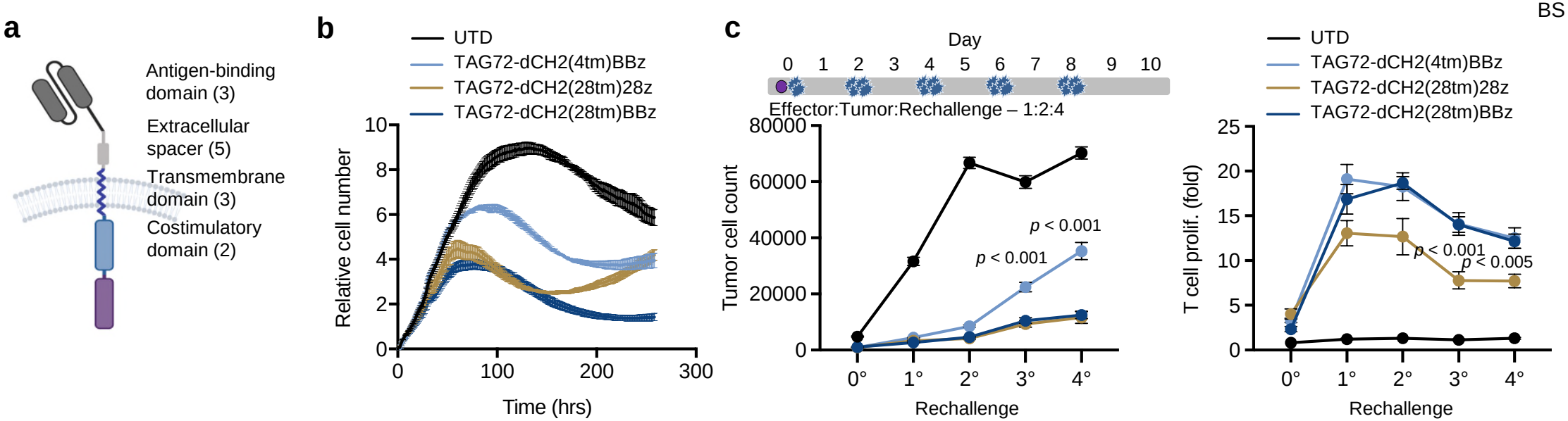
Building Better CARs: Optimizing the CAR construct



Eric Lee
BS



- TAG72-CARs with CD28 transmembrane domain with 4-1BB costimulation improves *in vitro* tumor cell killing and T cell expansion

Lee et al. *in revision*

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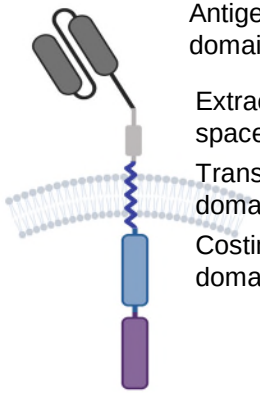
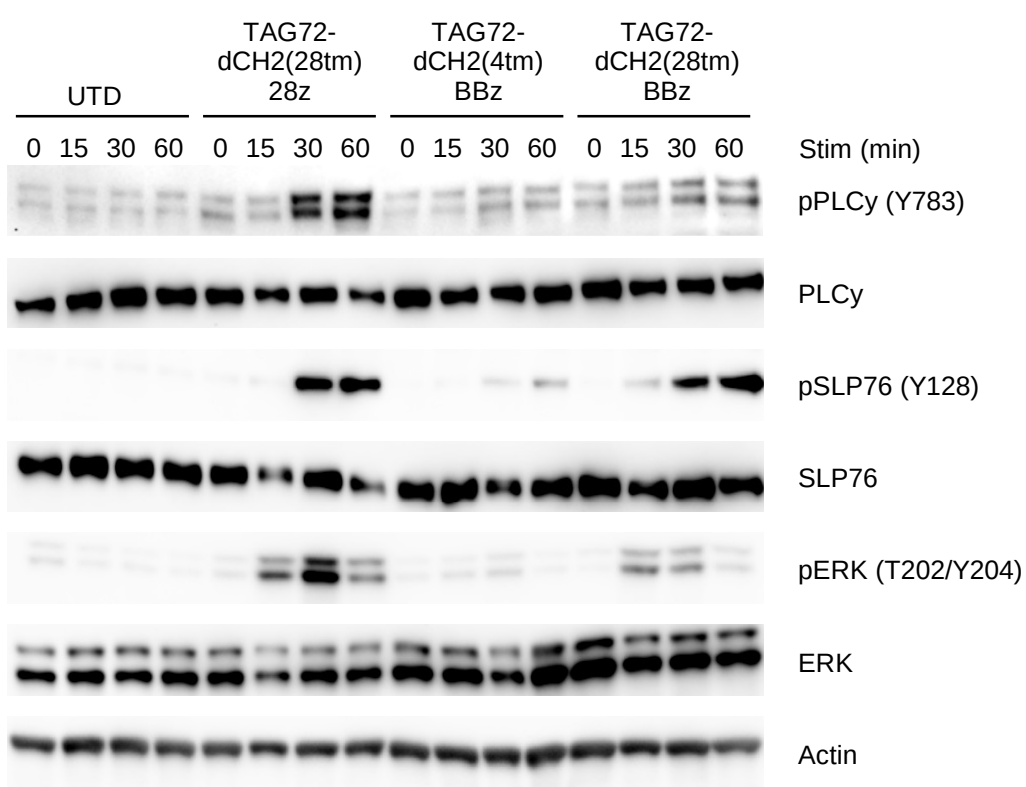
CD28 Transmembrane with 4-1BB Costimulation Shows Blended CD28/4-1BB Intracellular Signaling

a

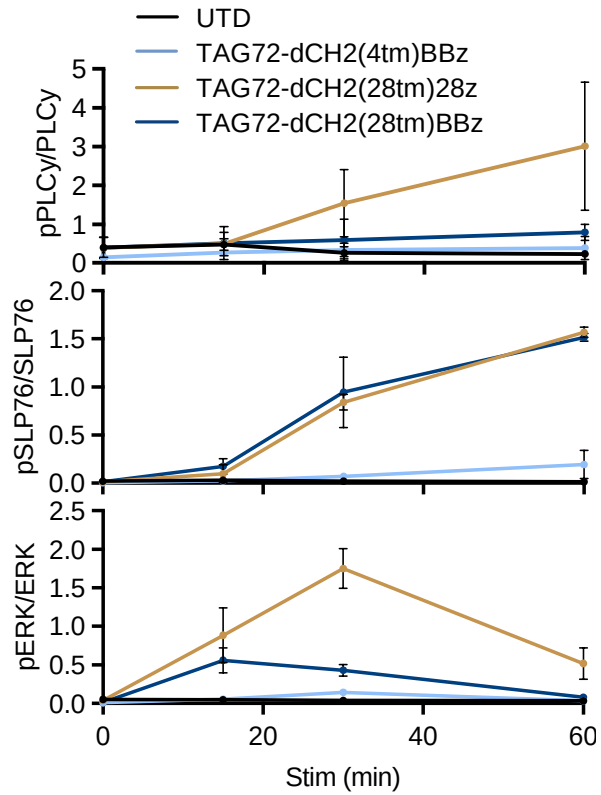


Collaboration with Leo Wang Lab:
Diana Gumber (Grad Student)

b



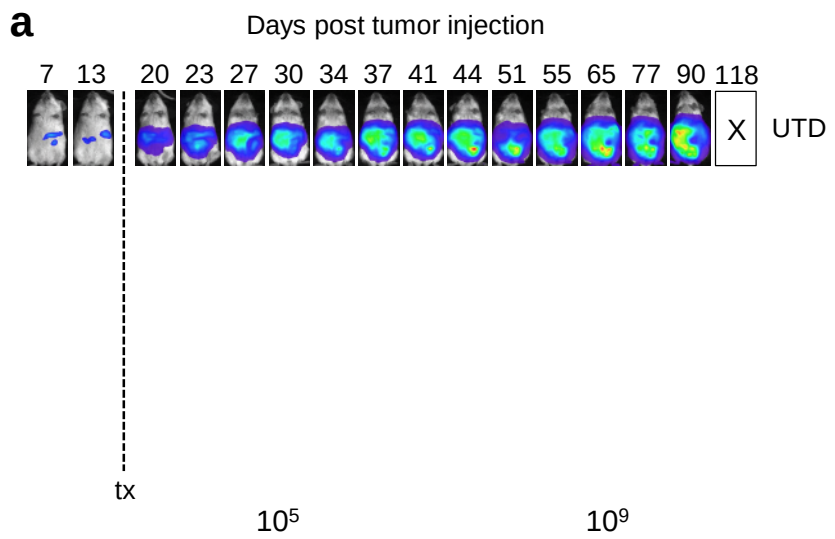
c

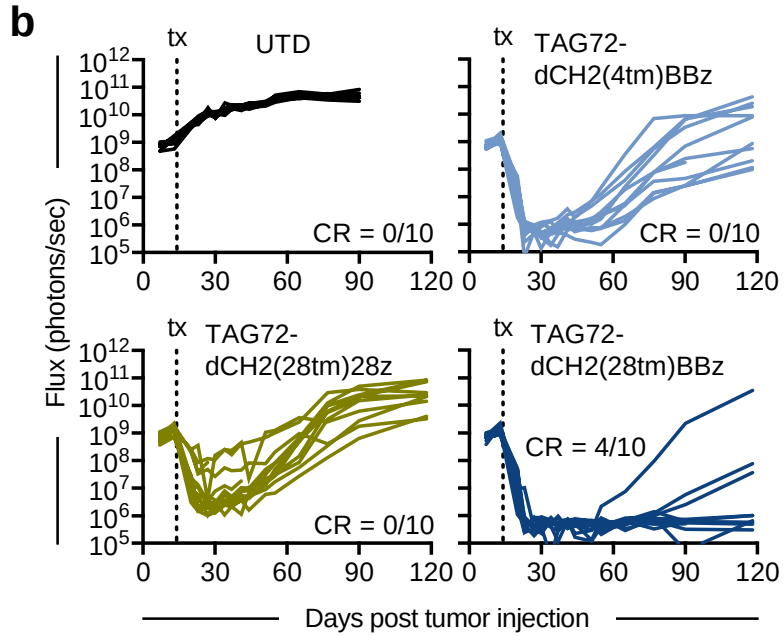


- TAG72-CARs with CD28 transmembrane domain with 4-1BB costimulation shows comparable pSLP76 but damped pPLCy and pERK signaling to CD28 costimulation

Lee et al. *in revision*

TAG72-28tmBBz CAR T Cells Provide Curative Responses against Ovarian Cancer Peritoneal Metastasis Xenograft Models

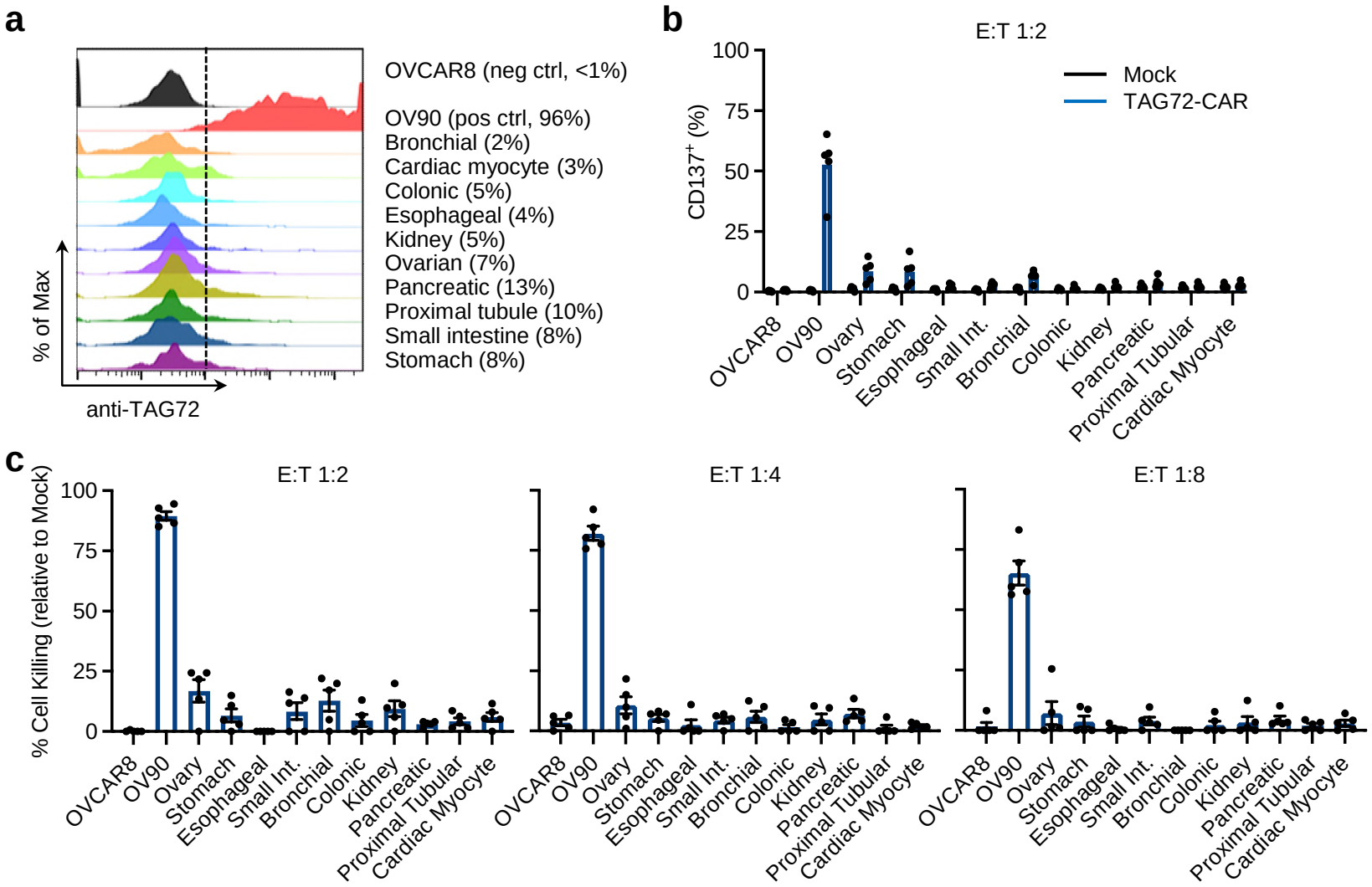




- TAG72-CARs with CD28 transmembrane domain with 4-1BB costimulation greatly improves *in vivo* anti-tumor efficacy

Lee et al. *in revision*

TAG72-28tmBBz CAR T Cells Demonstrate *In Vitro* Safety



- TAG72-CAR T cells show *in vitro* safety against primary human normal cells

Lee et al. *in revision*

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Phase 1 Clinical Trial to Evaluate TAG72-28tmBBζ CAR T Cells in Epithelial Ovarian Cancer

Lorna Rodriguez MD

- **TAG72+ platinum-resistant metastatic epithelial ovarian cancer**

  (Clinical PI: Lorna Rodriguez, MD PhD, Research PI: Saul Priceman, PhD) – Open to enrollment

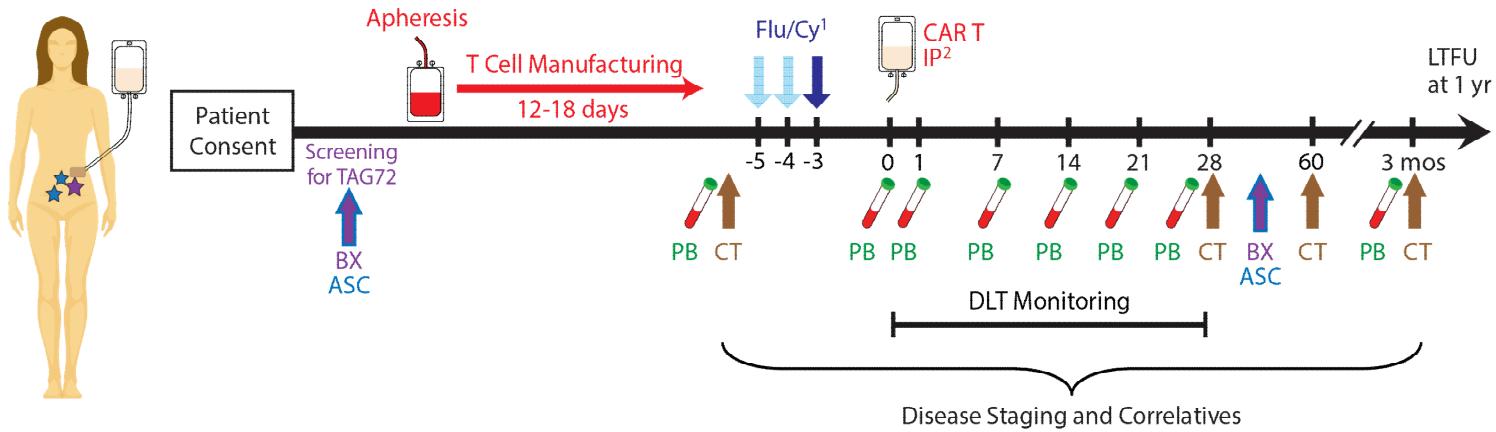

Table 1. CAR+ Cell Dose Schedule				
Dose -1	**Starting Dose 0a**	Dose 0b	Dose 1	Dose 2
50M	**100M**	100M +precond.	300M +precond.	600M + precond.

Murad et al. *Front Immunol* 2018
Lee et al. *in revision*

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Challenges Facing CAR T Cell Therapies for Solid Tumors

- Tumor antigen heterogeneity
- Immunosuppressive tumor microenvironment (TME)

Tethered IL-12: platform to increase T cell cytotoxicity of solid tumors and T cell expansion

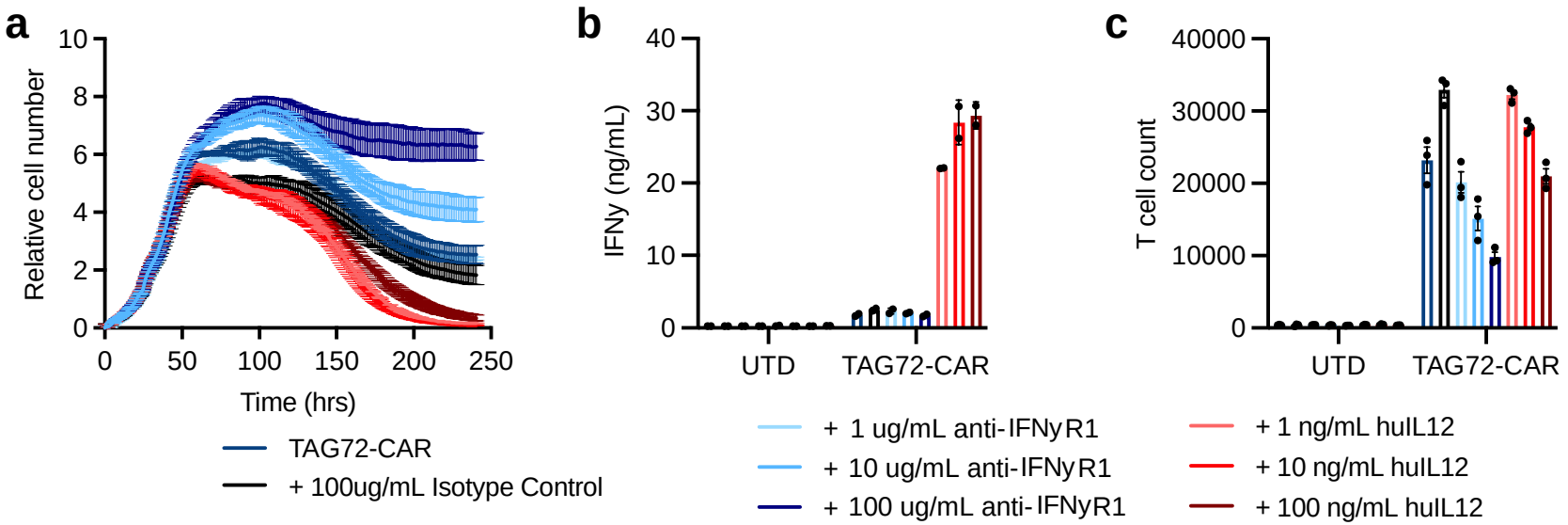
- The membrane-tethered IL12 molecule is a functional T cell intrinsic and tumor microenvironment modulating cytokine that we have shown potently increases T cell cytotoxicity of solid tumors, T cell expansion, and IFNg cytokine production of CAR T cells.
- We have also demonstrated that this molecule is antigen-dependent, and while it is constitutively made it is found largely intracellularly and non-functional until T cells are activated either by TCR or CAR stimulation. Upon stimulation, it is shuttled to the cell surface and enables downstream signaling.
- In preclinical mouse models, we show that engineering CAR T cells with membrane-tethered IL12 significantly improves anti-tumor responses and enhances the ability of CAR T cells to traffic to systemic sites of metastatic disease even from regional delivery (intraperitoneal for peritoneal metastasis disease models, and intracerebroventricular for brain metastatic disease models).
- We also demonstrate that engineering with membrane-tethered IL12 is a safe approach, compared with soluble injected IL12, using syngeneic mouse models.

IFNy is Critical for CAR T Cell Therapy for Solid Tumors

- Tumor eradication by CAR T cells relies on IFNg signaling on tumor stroma cells (Textor et al. *Cancer Res* 2014)
- IFNy is critical for TME effects in solid tumors through bystander effects (Thibaut et al. *Nat Cancer* 2020, Hoekstra et al. *Nat Cancer* 2020)
- Suppressive M2 macrophages targeted by the combination of CAR T cells and immune checkpoint blockade requires IFNg signaling (Yamaguchi et al. *JITC* 2022)
- CAR T cells modulate myeloid cells in GBM through IFNg (Alizadeh et al. *Cancer Discov* 2021)
- IFNg is critical for CAR T cell killing of solid tumors, but not liquid tumors (Larson et al. *Nature* 2022)

- **How do we endow CAR T cells with tumor-specific IFNg signaling?**

IL-12 Drives CAR T Cell Cytotoxicity via IFNg Signaling

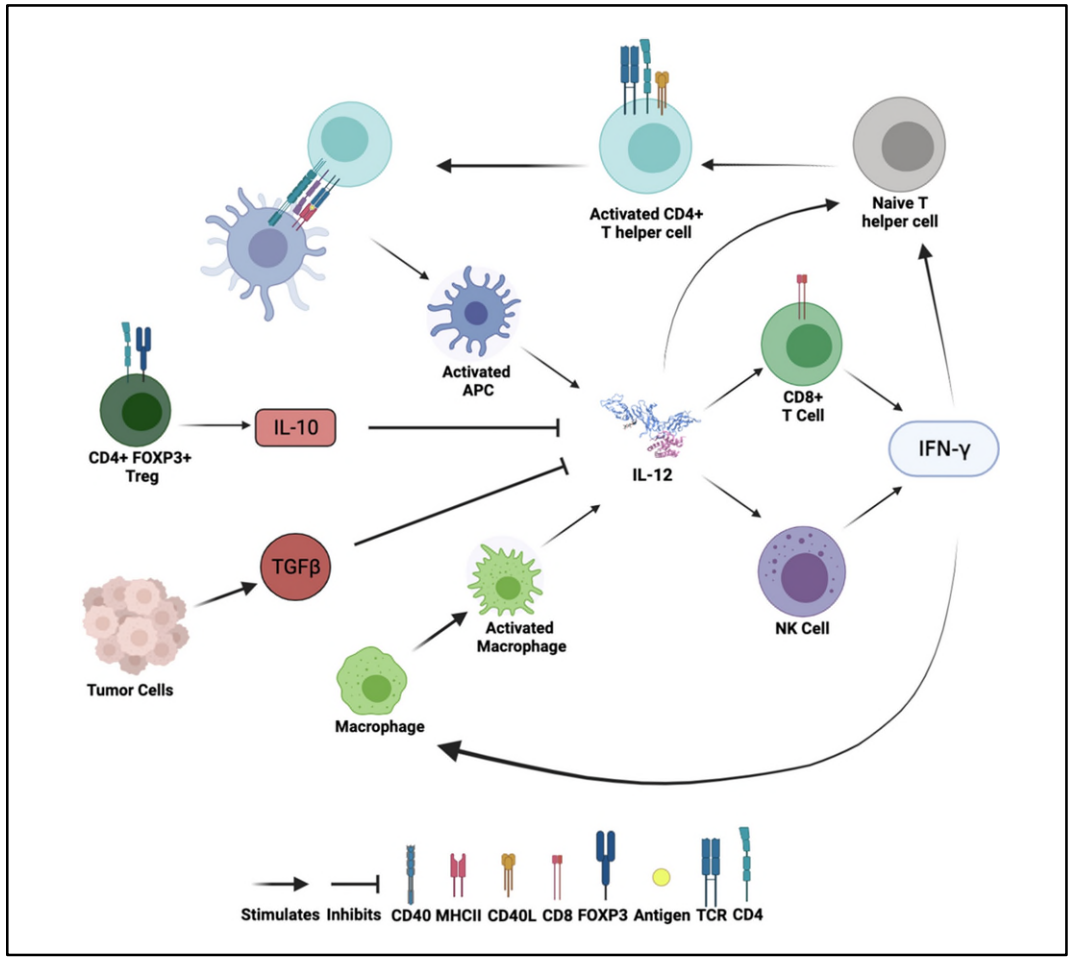


- IFNg is critical for tumor cell killing by TAG72-CAR T cells
- Exogenous IL-12 enhances tumor cell killing by TAG72-CAR T cells

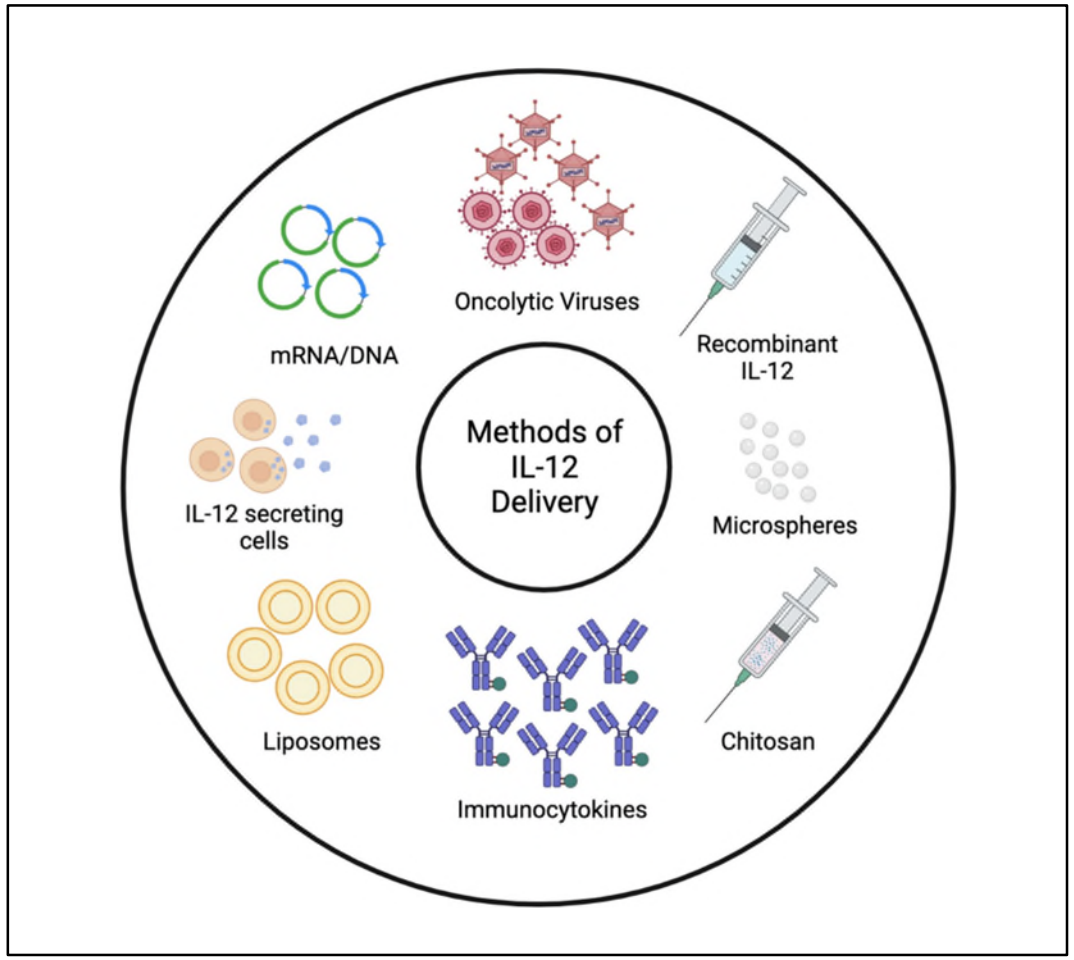
Lee et al. *in revision*

Overcoming TME Limitations on CAR T Cell Therapy: IL-12

Roles of IL-12 Signaling in the TME



How to safely deliver IL-12 to the TME?



Adapted from Cheng et al. *Cancer Immunol Immunother* 2022

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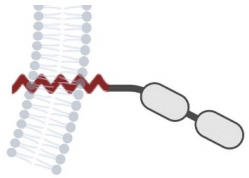
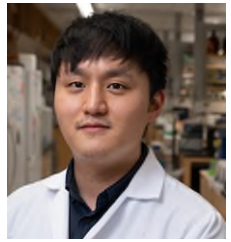
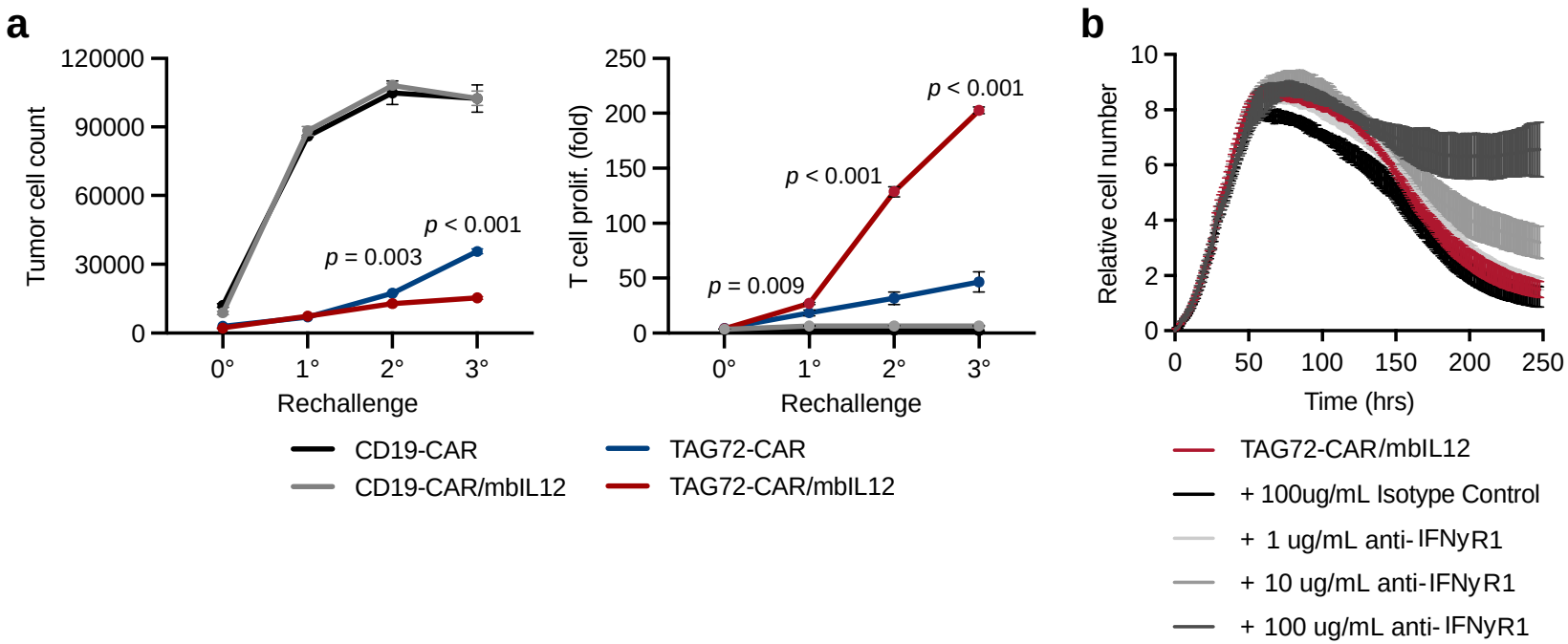
Engineering Membrane-Tethered IL-12 Signaling Drives IFNg-Mediated T Cell Cytotoxicity



mbIL12



Eric Lee
BS



- mbIL12 enhances T cell proliferation and tumor cell killing by TAG72-CAR T cells

Lee et al. *in revision*

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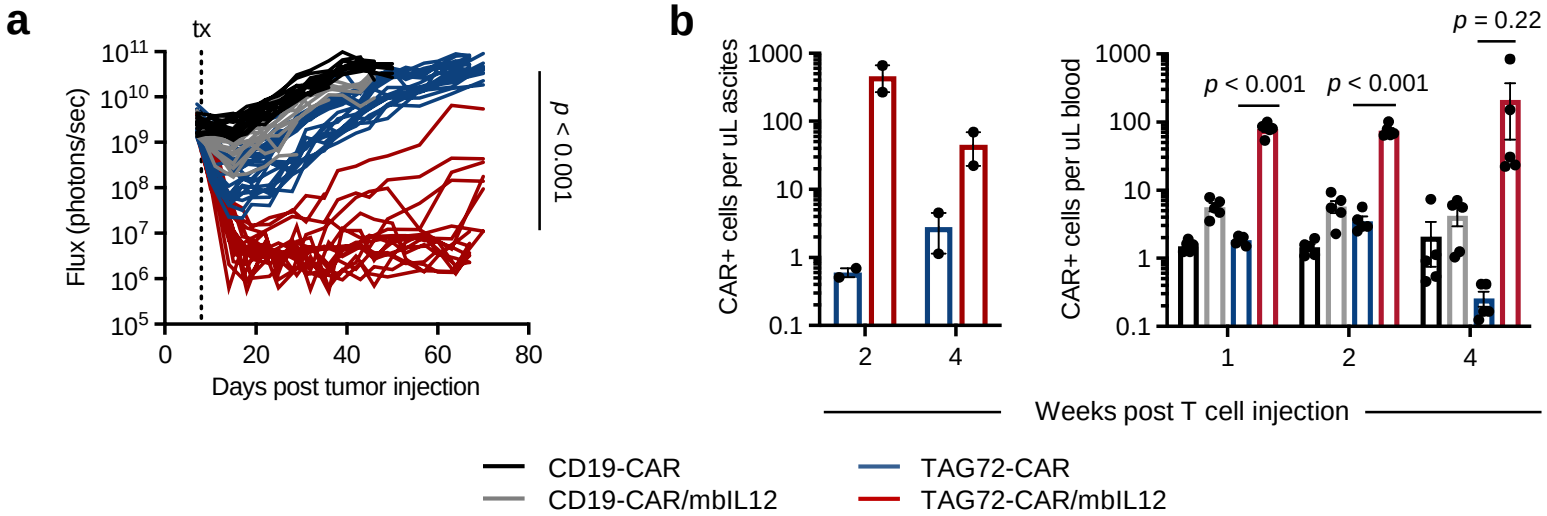
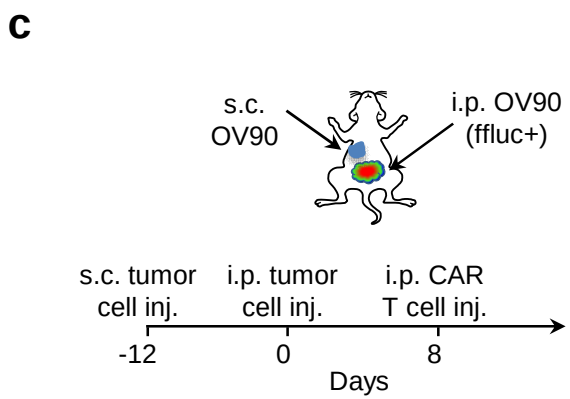
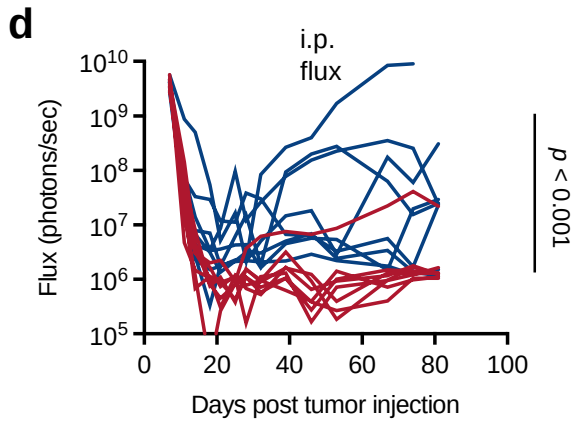
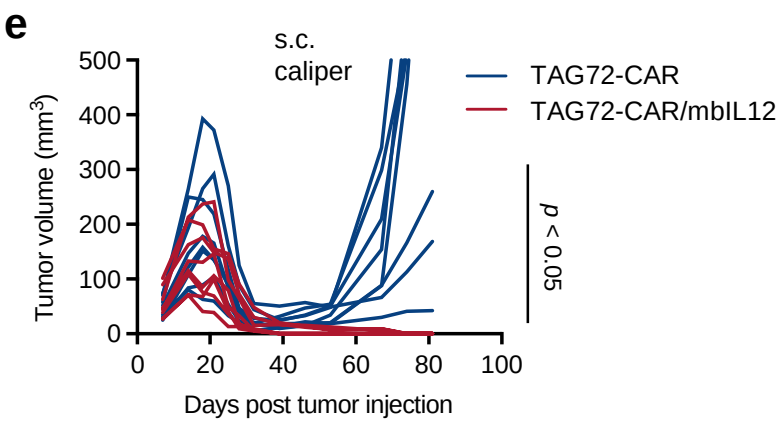
Membrane-Tethered IL-12 Signaling Promotes Anti-Tumor Efficacy and Regional-to-Systemic Disease Targeting









-Engineered mbIL12 cytokine signaling improves *in vivo* regional and systemic disease targeting

Lee et al. *in revision*

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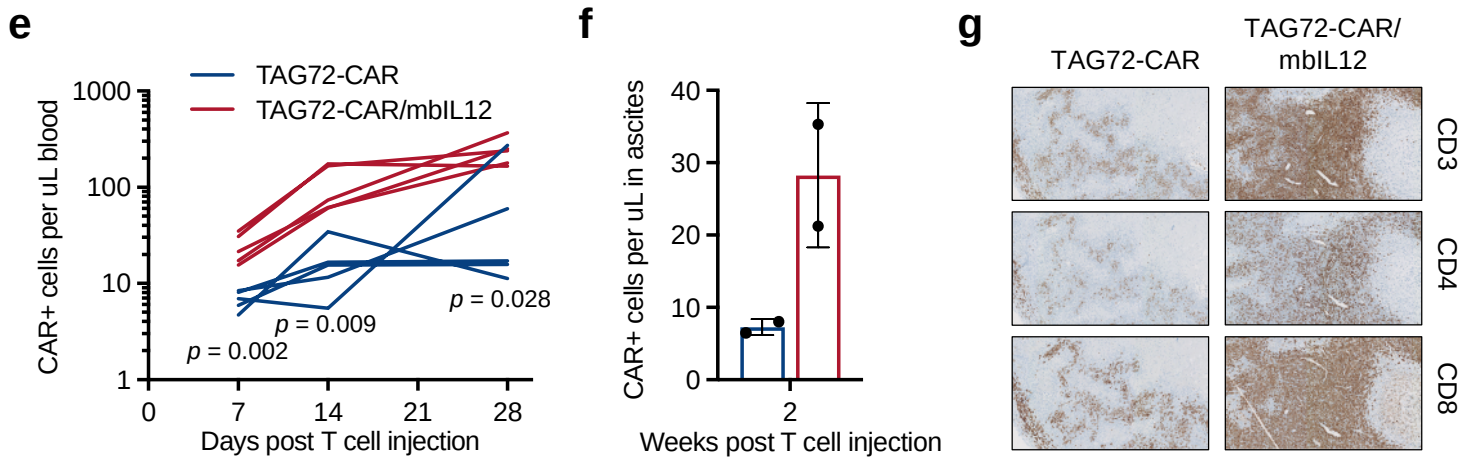
Membrane-Tethered IL-12 Signaling Promotes Anti-Tumor Efficacy and Regional-to-Systemic Disease Targeting



-Engineered mbIL12 cytokine signaling improves *in vivo* peripheral expansion and tumor infiltration regionally administered CAR T cells

Lee et al. *in revision*

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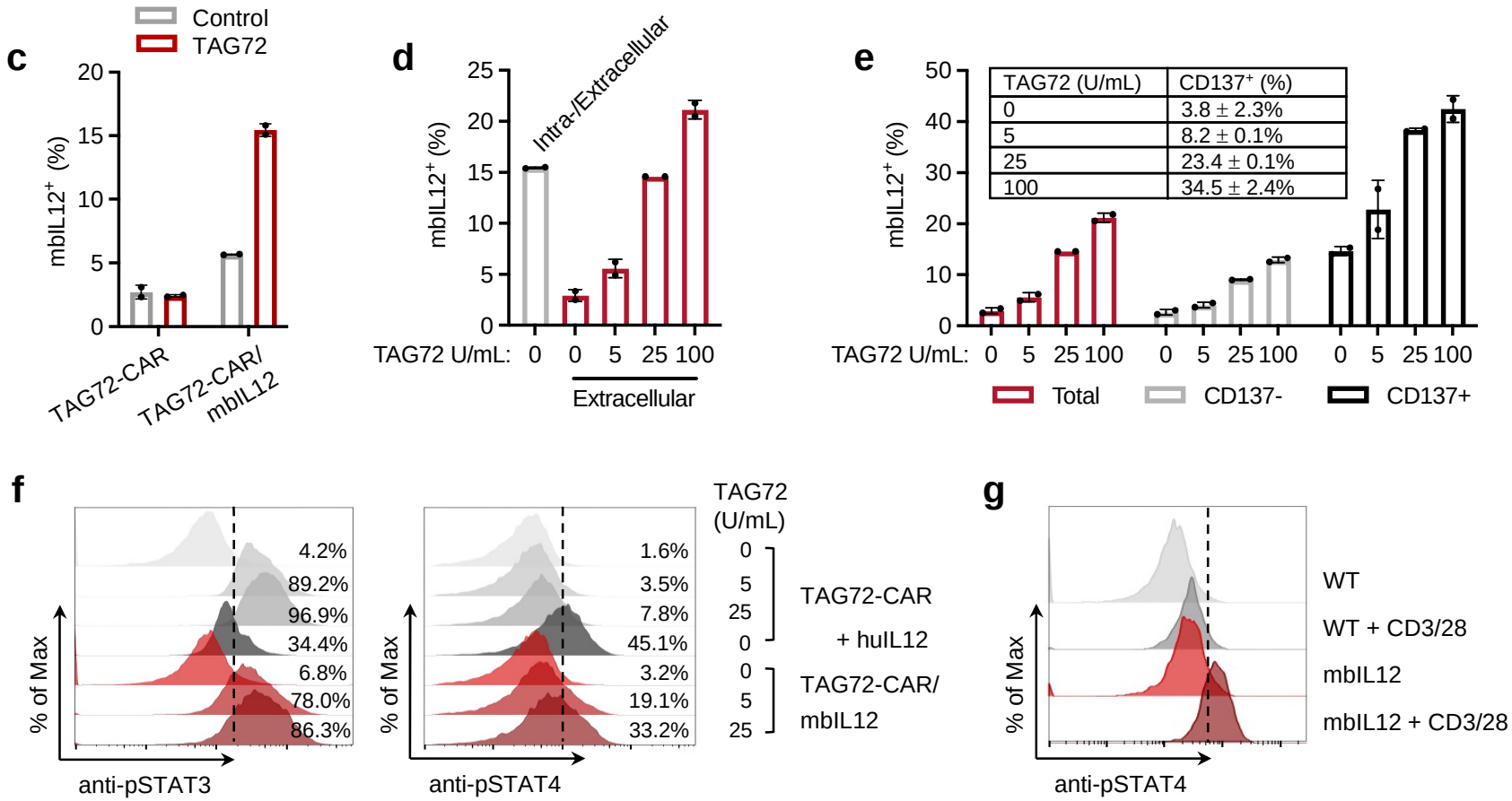
Membrane-Tethered IL-12 Signaling is Antigen-Dependent



- mbIL12 is shuttled to cell surface and induces downstream signaling only after T cell activation
- mbIL12 can signal in *cis* and in *trans*

Lee et al. *in revision*

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CONTACT INFORMATION

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