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# CAR-T Cell Therapies: From Origin to Acceleration

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**Center for Cellular Immunotherapies, University of Pennsylvania**

**Co-Founder, Tmunity Therapeutics**

**President, International Society for Cell and Gene Therapy**

**@BLLPHD**

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# Conflict of Interest Statement

- Declaration of financial interest due to intellectual property and patents in the field of cell and gene therapy.
- University of Pennsylvania Alliance with Novartis
- Consultant for Terumo
- Scientific Advisory Board for Akron, Avectas, Immuneel, Immusoft, In8bio, Ori Biotech, Vycellix
- Co-Founder and equity holder Tmunity Therapeutics
- Conflict of interest is managed in accordance with University of Pennsylvania policy and oversight

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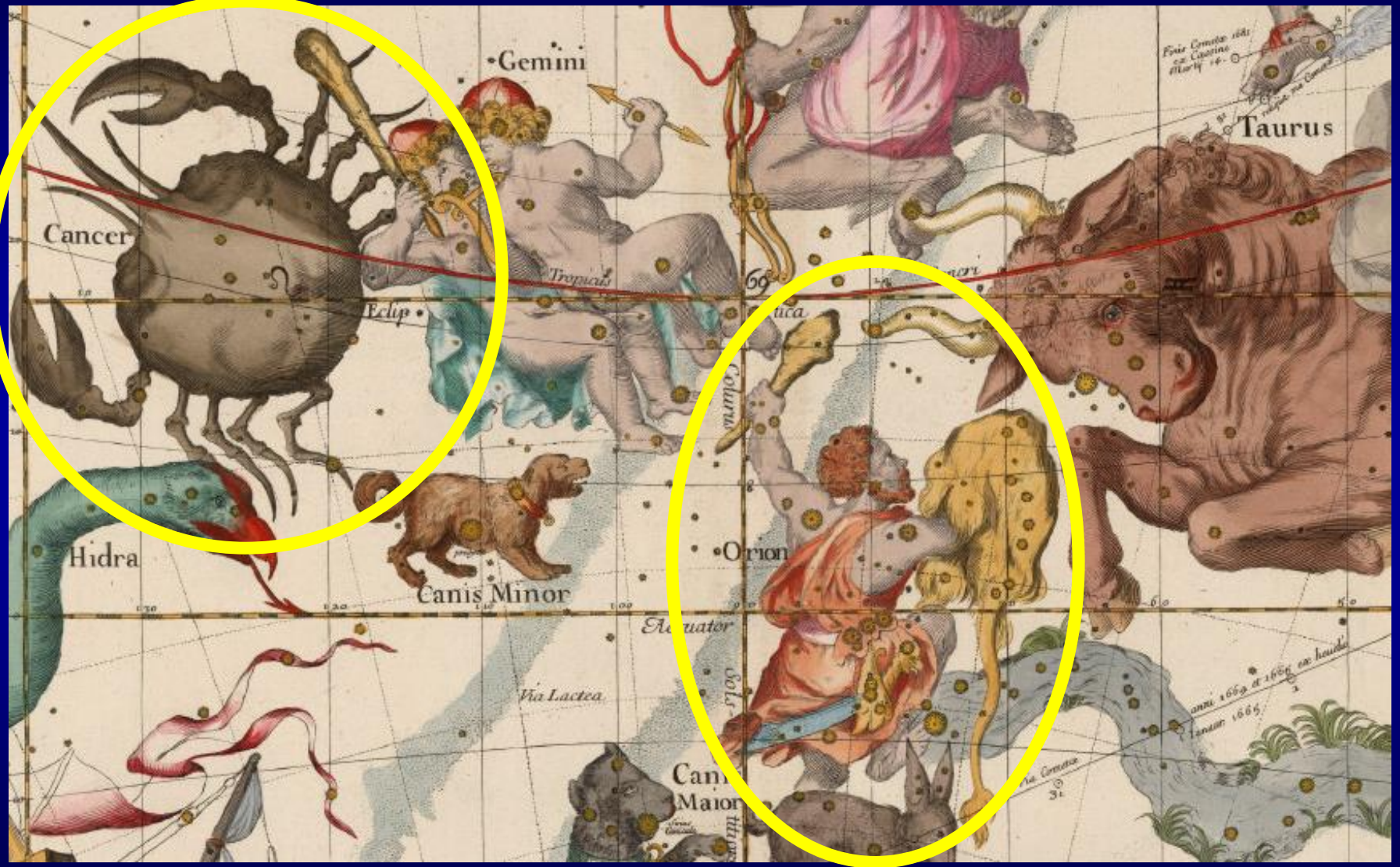
# Redirecting Orion to Hunt Cancer




Ignace-Gaston Pardies, Star Atlas 2nd edition 1693, Paris

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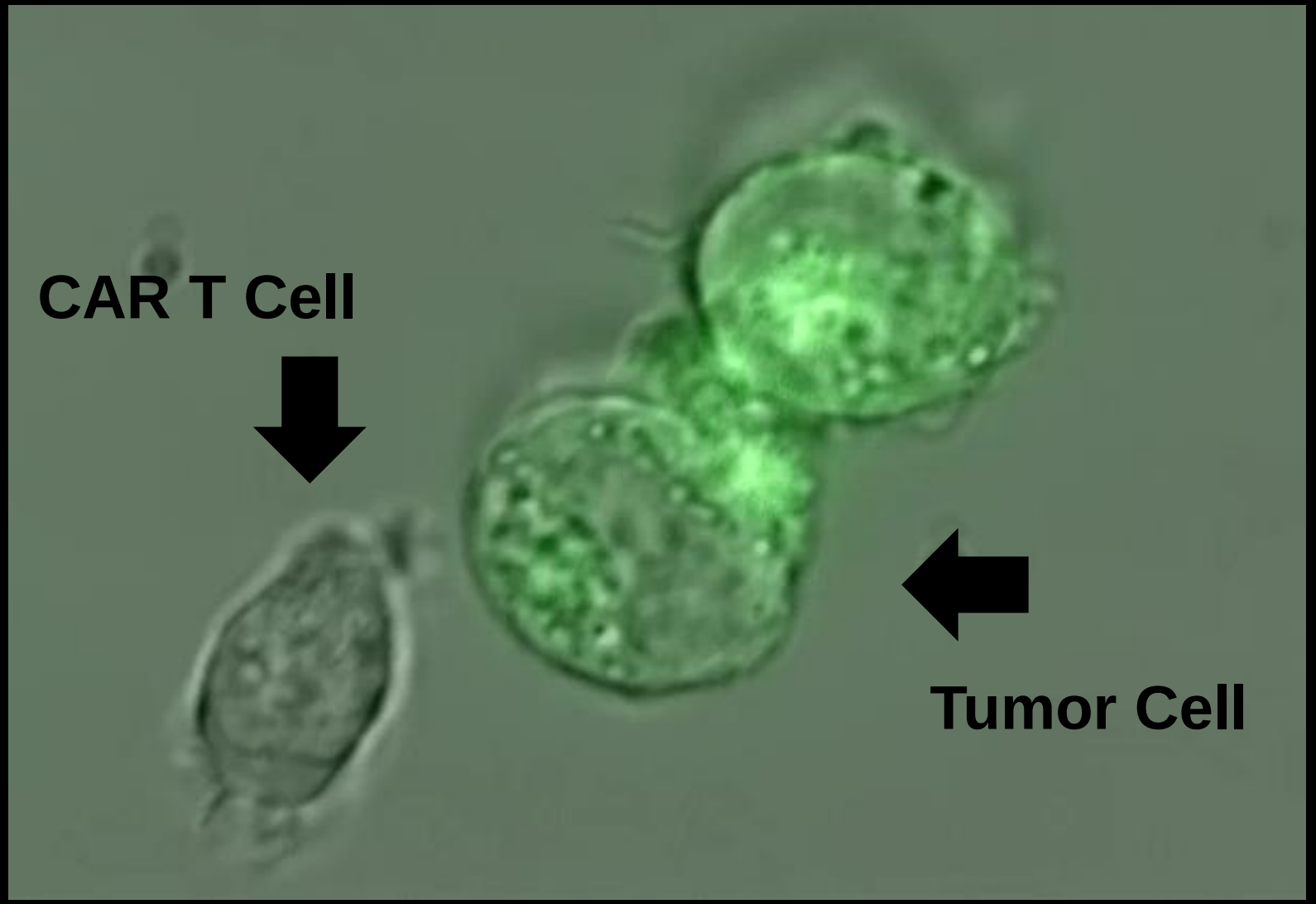
# Engineered Immunity:
# Chimeric Antigen Receptor (CAR) T Cells To Kill Cancer



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# Redirection of Specificity and Effector Function: The Beginning

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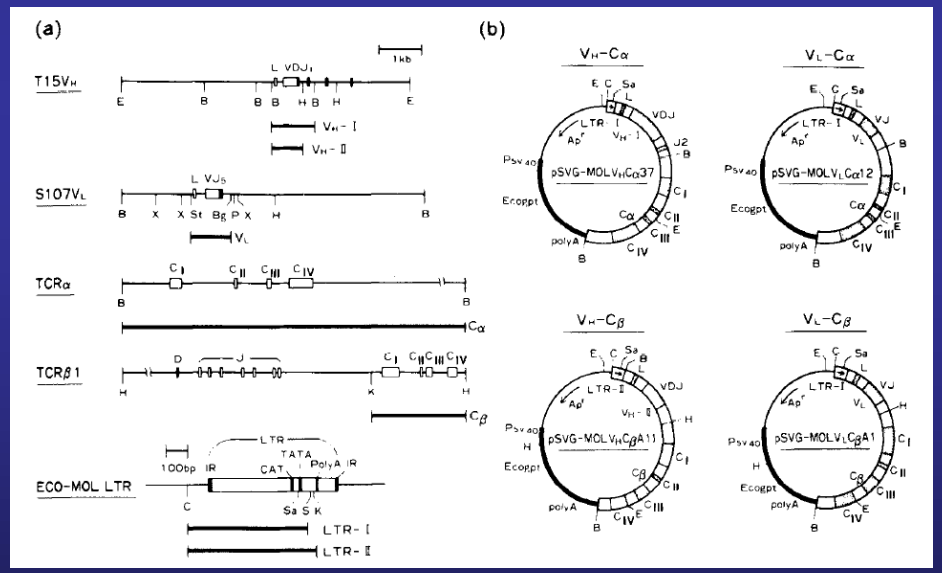
# B Cell Receptors + T Cell Receptors: 1987-1989

Vol. 149, No. 3, 1987
December 31, 1987
BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS
Pages 960-968

EXPRESSION OF CHIMERIC RECEPTOR COMPOSED OF IMMUNOGLOBULIN-DERIVED V REGIONS AND T-CELL RECEPTOR-DERIVED C REGIONS

Yoshihisa Kuwana[1], Yoshihiro Asakura[1], Naoko Utsunomiya[2], Mamoru Nakanishi[2], Yohji Arata[2], Seiga Itoh[3], Fumihiko Nagase[4] and Yoshikazu Kurosawa[1*]




In future, it might become possible for T cells recognizing any antigens without MHC restriction to be produced by the technique described in this paper.

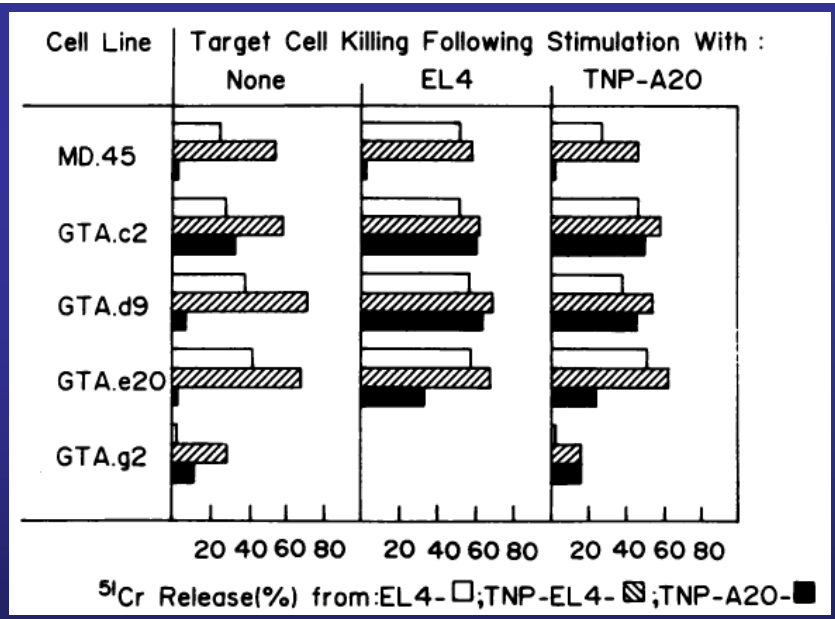
*Proc. Natl. Acad. Sci. USA*
Vol. 86, pp. 10024–10028, December 1989
Immunology

## Expression of immunoglobulin-T-cell receptor chimeric molecules as functional receptors with antibody-type specificity

(chimeric genes/antibody variable region)

GIDEON GROSS, TOVA WAKS, AND ZELIG ESHHAR*

Department of Chemical Immunology, The Weizmann Institute of Science, Rehovot 76100, Israel




This approach can be exploited, for example, to direct cytotoxic T lymphocytes to kill tumor or virally infected cells.

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# First CAR Publications in Academia
# First CAR Clinical Trials in Industry

CELL GENESYS

**RECOMBINANT DNA ADVISORY COMMITTEE MEETING**
**March 3-4, 1994**
**CD4-zeta in HIV+ Subjects**

## Long-term in vivo survival of receptor-modified syngeneic T cells in patients with human immunodeficiency virus infection

Robert E. Walker, Christine M. Bechtel, Ven Natarajan, Michael Baseler, Kristen M. Hege, Julia A. Metcalf, Randy Stevens, Allison Hazen, R. Michael Blaese, Clara C. Chen, Susan F. Leitman, Jolie Palensky, Janet Wittes, Richard T. Davey Jr, Judith Falloon, Michael A. Polis, Joseph A. Kovacs, David F. Broad, Bruce L. Levine, Margo R. Roberts, Henry Masur, and H. Clifford Lane

BLOOD, 15 JULY 2000 VOLUME 96, NUMBER 2

## Prolonged survival and tissue trafficking following adoptive transfer of CD4ζ gene-modified autologous $CD4^+$ and $CD8^+$ T cells in human immunodeficiency virus–infected subjects

Ronald T. Mitsuyasu, Peter A. Anton, Steven G. Deeks, David T. Scadden, Elizabeth Connick, Matthew T. Downs, Andreas Bakker, Margo R. Roberts, Carl H. June, Sayeh Jalali, Andy A. Lin, Rukmini Pennathur-Das, and Kristen M. Hege

BLOOD, 1 AUGUST 2000 • VOLUME 96, NUMBER 3

Hege et al. Journal for ImmunoTherapy of Cancer (2017) 5:22
DOI 10.1186/s40425-017-0222-9

Journal for ImmunoTherapy of Cancer

RESEARCH ARTICLE Open Access

## Safety, tumor trafficking and immunogenicity of chimeric antigen receptor (CAR)-T cells specific for TAG-72 in colorectal cancer

Kristen M. Hege[1,2,3*], Emily K. Bergsland[3], George A. Fisher[4], John J. Nemunaitis[5], Robert S. Warren[3], James G. McArthur[1], Andy A. Lin[1], Jeffrey Schlom[6], Carl H. June[7] and Stephen A. Sherwin[1,3]

**First CAR T Trial in Cancer**

**Conducted 1997**
**Published 2017!**

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# Women of Early CAR T Cell Development

**Helene Finney**

- CellTech (UCB)
- Chimeric receptors providing both primary and costimulatory signaling in T cells from a single gene product. Journal of Immunology. 1998;161:2791-7

**Kristen Hege**

- Cell Genesys
- Celgene
- Led first ever clinical trial of CAR T Cells in cancer 1997

**Margo Roberts**

- Cell Genesys
- Kite
- Chimeric receptor molecules for delivery of co-stimulatory signals US Patent 5,686,281 filed May 31, 1995, granted November 11, 1997

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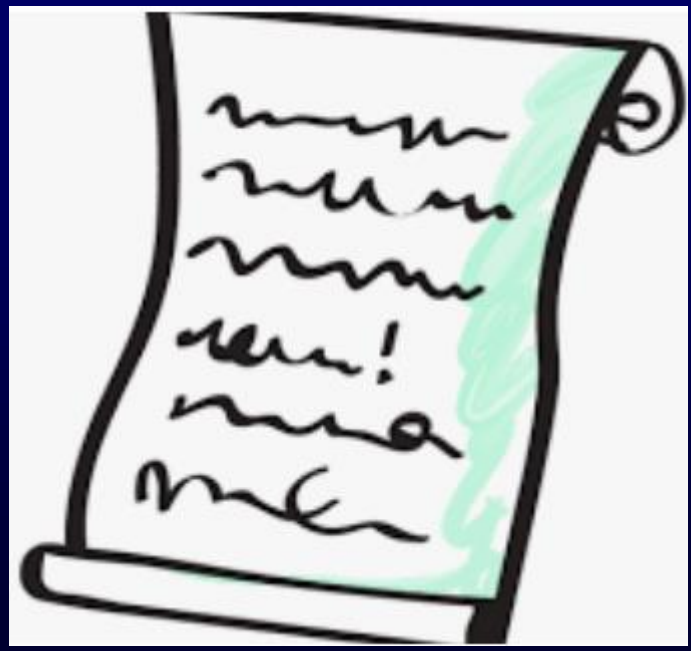
# Agenda

- CAR T cells in Liquid Tumors
- CAR T cells in Solid Tumors
- Multiplex Engineering
- Where are We Going?



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# August, 2010
# CAR T Cells Targeting CD19 in CLL

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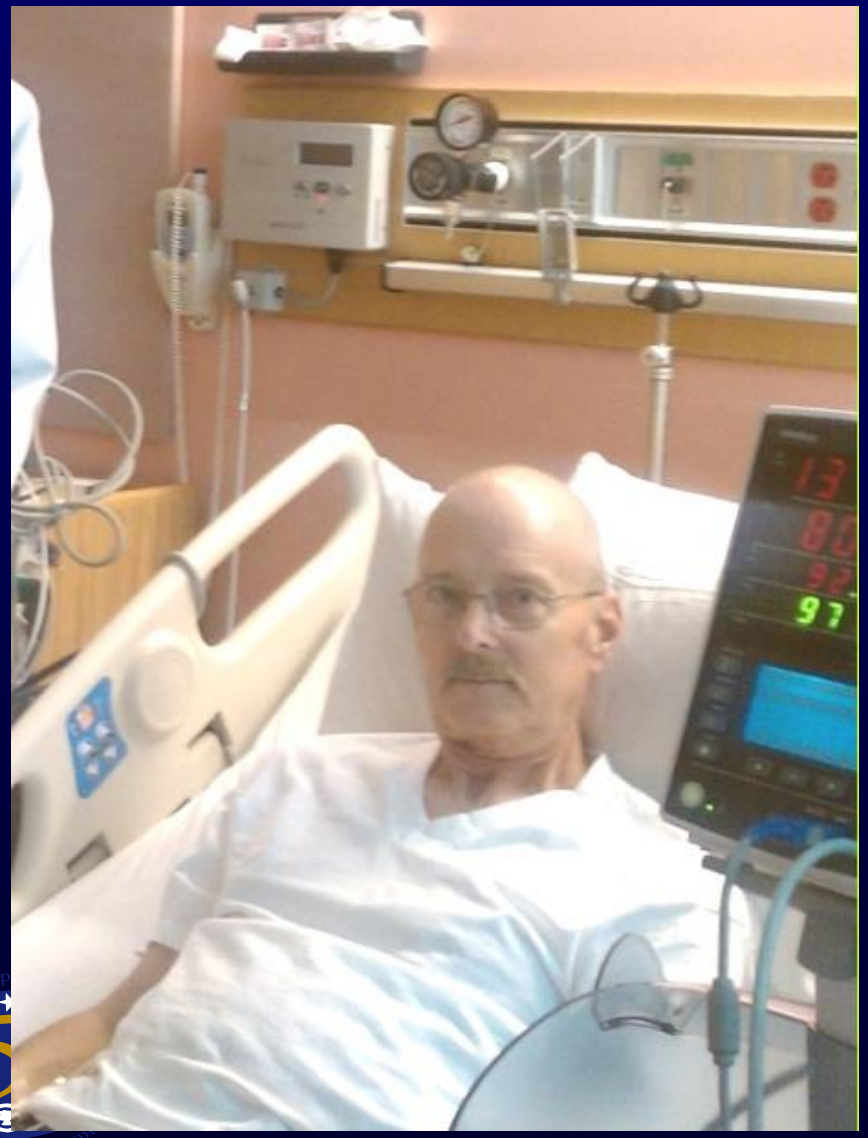
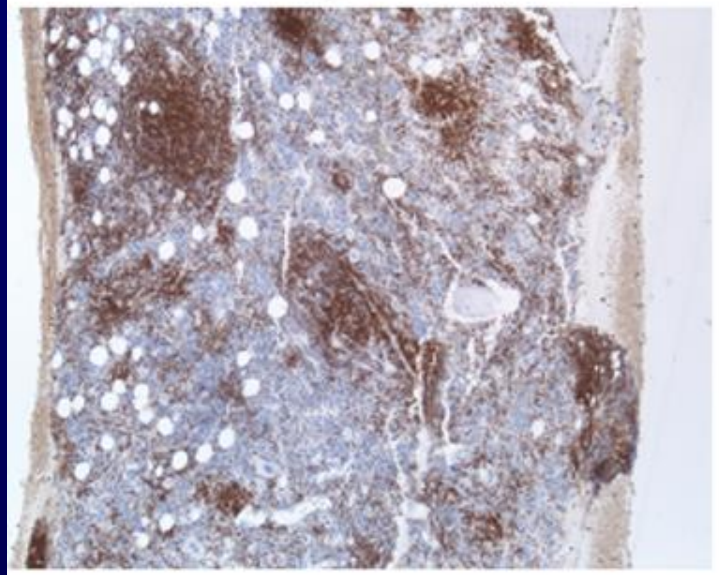
# August, 2010



Before CAR T Cells



**Pt #1**

**2.9 (1.3)**

**Pt#2**

**5.5 (2.5)**

**Pt #3**

**7.7 (3.5)**

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# April, 2012
# CAR T Cells Targeting CD19 in ALL

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# Global Regulatory Approvals

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NHK WORLD-JAPAN > News > Japan > Japan approves new cancer immunotherapy

Japan

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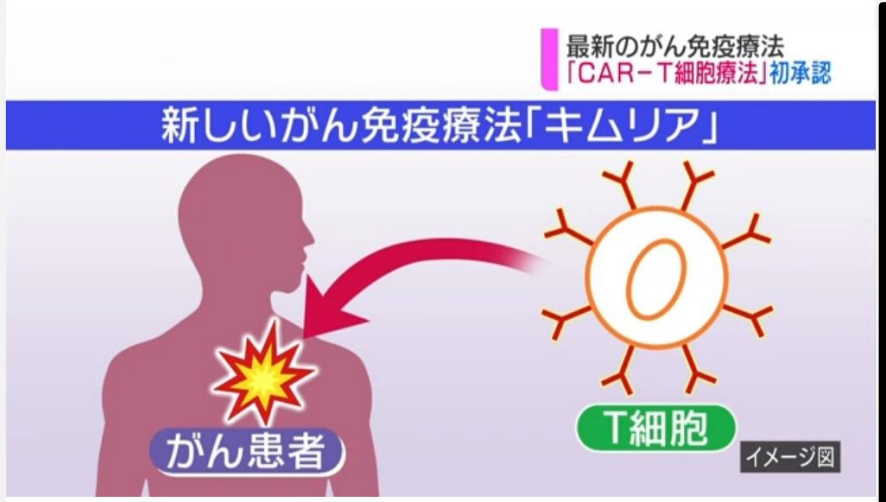
## Japan approves new cancer immunotherapy

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- Researchers test drug for treating ALS

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The Sydney Morning Herald

EXCLUSIVE NATIONAL CANCER

## 'Revolutionary' cancer drug using genetically modified cells approved

By Esther Han
December 19, 2018 — 12.00am

A revolutionary cancer therapy that supercharges a patient's immune cells to hunt and destroy cancer cells has been approved for use in Australia, ushering in a new era in medicine.

Desperate patients with aggressive blood cancers who have not responded to conventional treatments have been heading overseas to receive a shot of the "custom-made" drug and

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The Journal of Clinical Investigation RESEARCH ARTICLE

# CD19-targeting CAR T cell immunotherapy outcomes correlate with genomic modification by vector integration

Christopher L. Nobles,[1] Scott Sherrill-Mix,[1] John K. Everett,[1] Shantan Reddy,[1] Joseph A. Fraietta,[1,2,3,4,5] David L. Porter,[2,4,6] Noelle Frey,[2,4,7] Saar I. Gill,[2,4,7] Stephan A. Grupp,[6] Shannon L. Maude,[6] Donald L. Siegel,[2,3] Bruce L. Levine,[2,3,4] Carl H. June,[2,3,4,5] Simon F. Lacey,[2,3,4] J. Joseph Melenhorst,[2,3,4] and Frederic D. Bushman[1]

[1]Department of Microbiology, [2]Center for Cellular Immunotherapies, [3]Department of Pathology and Laboratory Medicine, and [4]Abramson Cancer Center, Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, USA. [5]Parker Institute for Cancer Immunotherapy, University of Pennsylvania, Philadelphia, Pennsylvania, USA. [6]Division of Oncology, The Children's Hospital of Philadelphia, Philadelphia, Pennsylvania, USA. [7]Division of Hematology-Oncology, Department of Medicine, Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, USA.

J Clin Invest. 2020 Feb 3;130(2):673-685.

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# Outstanding Fate Mapping Questions

- Do you “get what you get?”
- Should you select (patients or cells) and how do we know we are not missing important cells or denying patients?
- Passengers or Drivers?
- Experimentation and Iteration

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# Kymriah for CLL: Summary

- A subset of patients with refractory CLL have durable remissions after Kymriah
- For refractory CLL, the CR rate with Kymriah is about ~30%
  - Frey NV et al. J Clin Oncol. 2020;38(25):2862-71
- CLL cells induce CAR T dysfunction
  - Singh N et al. Cancer Discov. 2020;10(4):552-67
- BTK/ITK inhibitors appear to synergize with CD19 CAR T
  - Turtle CJ. J Clin Oncol. 2017;35(26):3010-20, Gill, S. et al, unpublished
- CLL is emblematic of the tug-of-war between effective but non-curative targeted reagents (ibrutinib, venetoclax, etc) and a potentially one-time curative T cell-based therapy!

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# Solid Tumor Challenges

- Antigen Coverage
- Antigen Escape
- Antigen Specificity
- Immunosuppressive Microenvironment
- Trafficking

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# Solid Tumor Challenges

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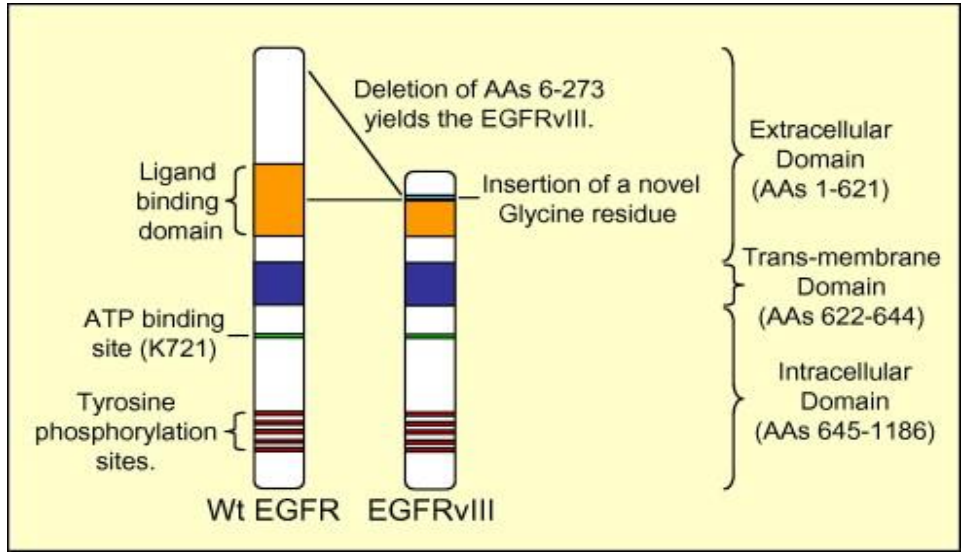
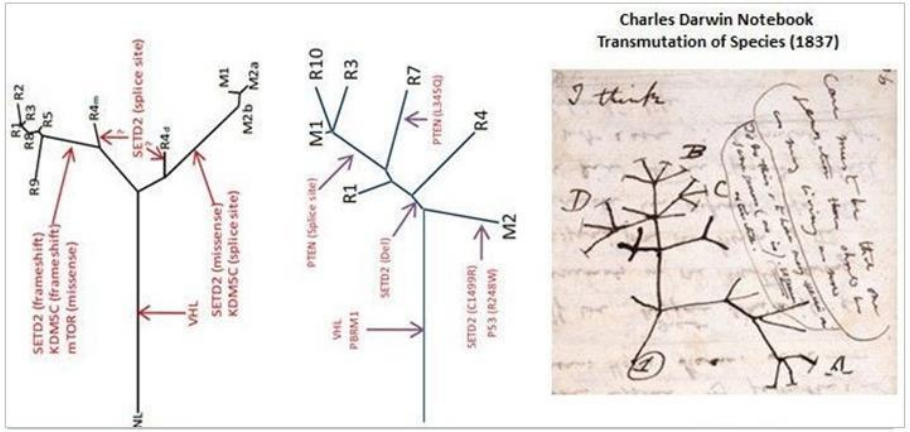
# EGFRviii as a CAR Target for GBM

## Pros and Cons as a CAR Target

- Pro: tumor specific mutation so that on-target, off-tumor toxicity is unlikely
- Con: EGFRviii is a subclonal mutation: tumor heterogeneity




- Francis JM. EGFR variant heterogeneity in glioblastoma resolved through single-nucleus sequencing. *Cancer discovery. 2014;4(8):956-71.*

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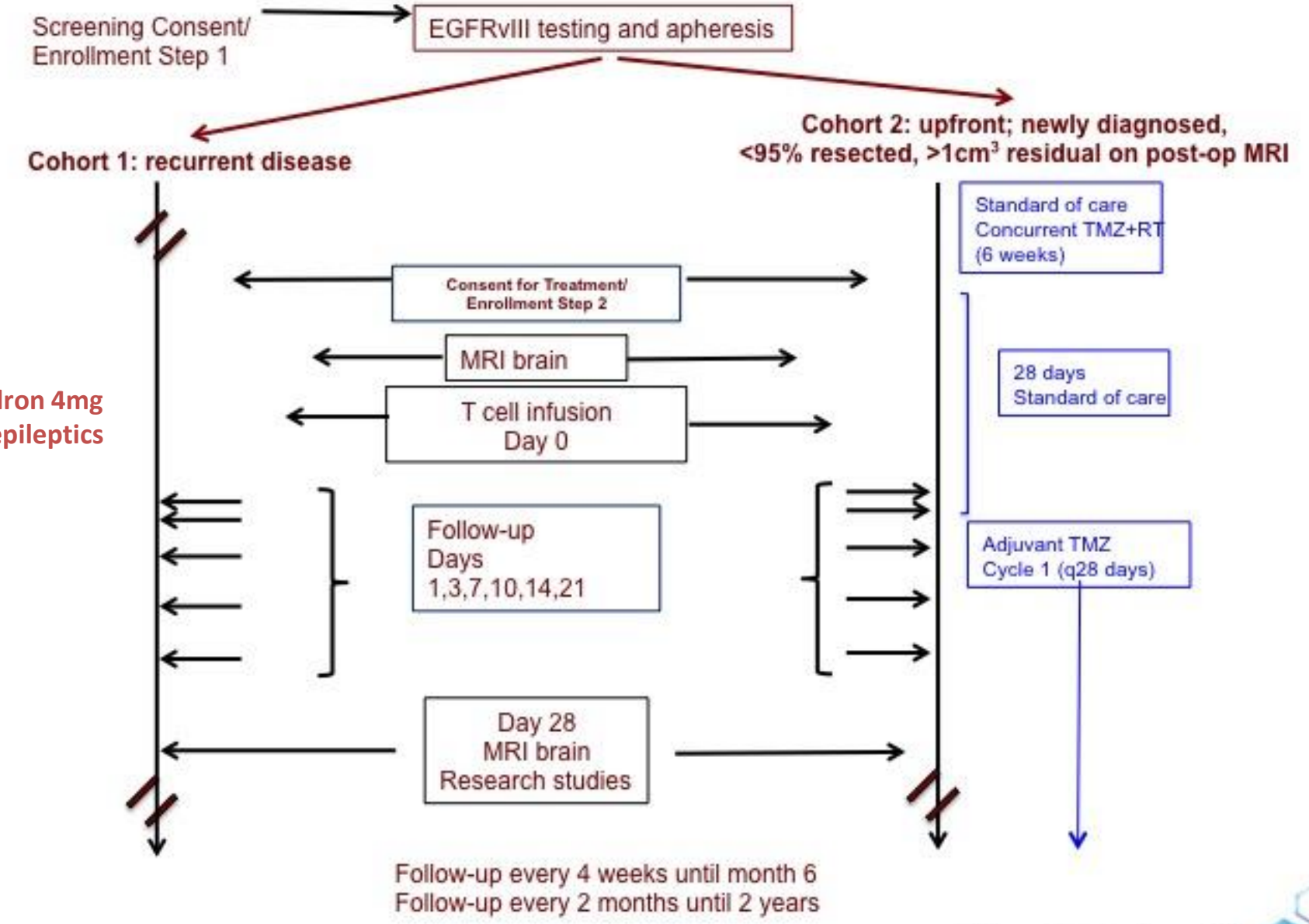
# Protocol schema: NCT02209376



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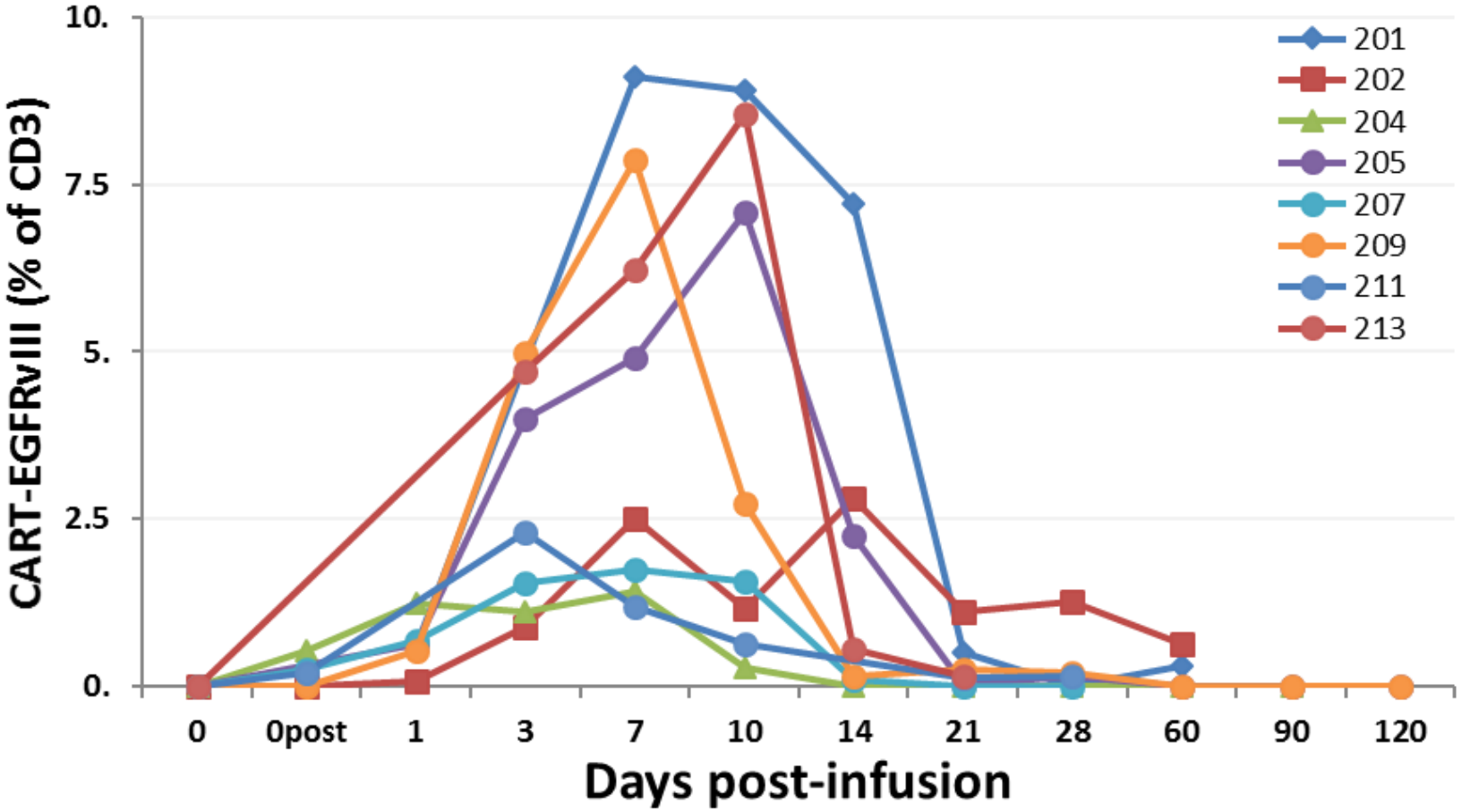
# CART engraftment and persistence in blood

## Flow Cytometry




A single dose of peripherally infused EGFRvIII-directed CAR T cells mediates antigen loss and induces adaptive resistance in patients with recurrent glioblastoma, O'Rourke, D.M., et al. Sci Transl Med. 2017 Jul 19;9(399):eaaa0984

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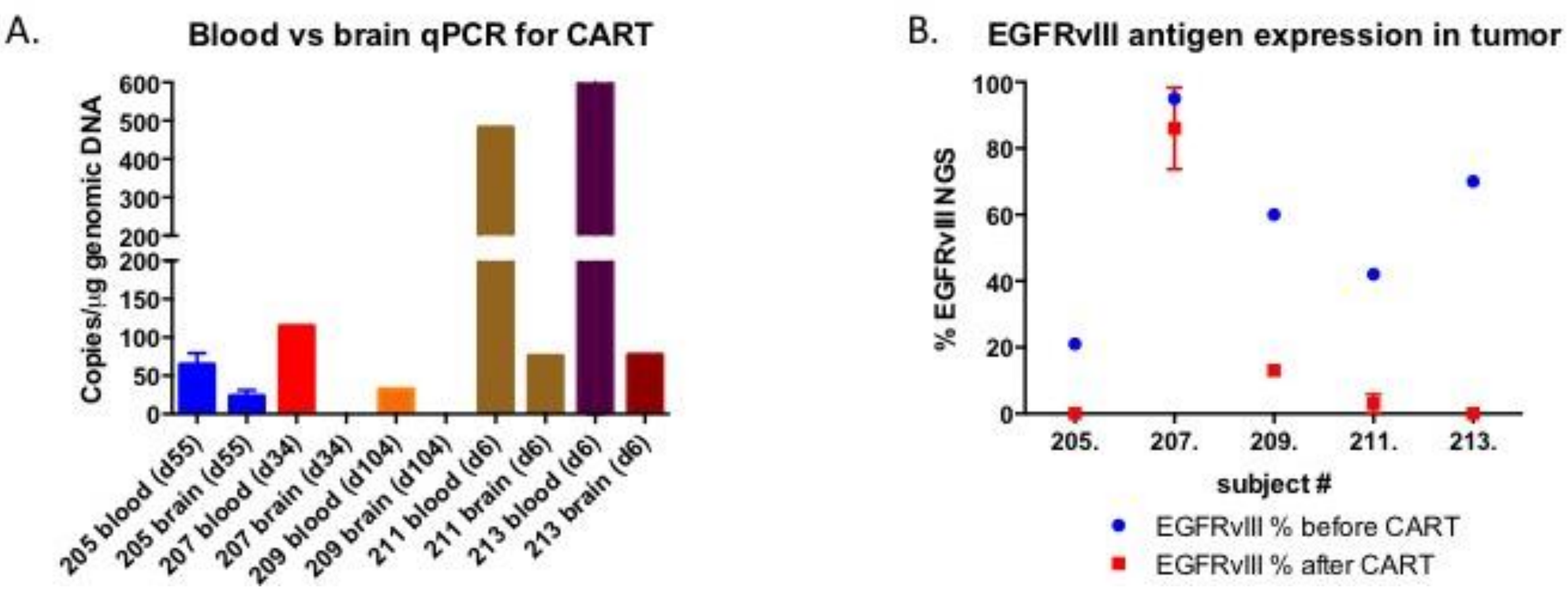
# CART Trafficking to Brain and Downregulation of EGFRviii




**A single dose of peripherally infused EGFRvIII-directed CAR T cells mediates antigen loss and induces adaptive resistance in patients with recurrent glioblastoma, O'Rourke, D.M., et al. Sci Transl Med. 2017 Jul 19;9(399):eaaa0984**

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# CART EGFRvIII in Glioblastoma Conclusions

- Screening for EGFRvIII and CART-EGFRvIII manufacturing is feasible for GBM patients
- Infusion of CART-EGFRvIII is safe, but may induce seizures.
- There is no EGFR-directed toxicity in 8 of 8 patients (not cetuximab!)
- Mechanism of Action established:
  - CART-EGFRvIII expand in blood and traffic to brain.
  - CART-EGFRvIII detectable (Q-PCR analysis) in brain tumor in 3 of 5 patients
  - EGFRvIII antigen loss (NGS analysis) in brain tumor in 4 of 5 patients
- Induction of new T cell infiltrates by IHC in tumor resection specimens may suggest antigenic spreading or bystander T cells
- Additional strategies: target other antigens to prevent tumor escape, combine with checkpoint Ab

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# Solid Tumor Challenges

- Antigen Coverage
- Antigen Escape
- Antigen Specificity
- Immunosuppressive Microenvironment
- Trafficking

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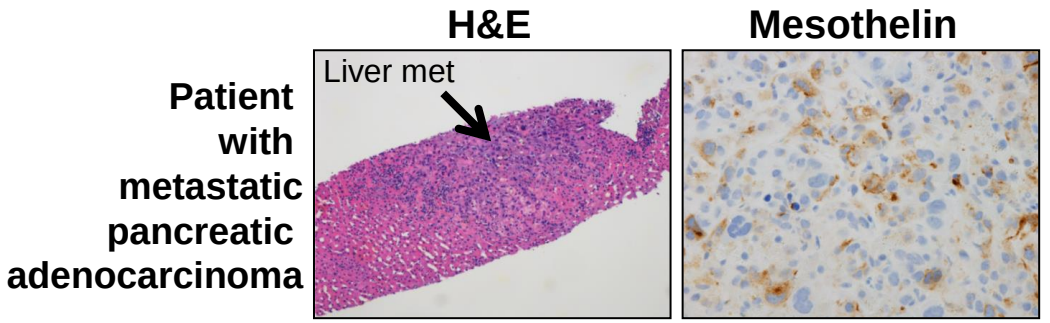
# Mesothelin as a Target for CAR T cells

## Description and characterization

- GPI anchored membrane protein (40 kDa) that can be shed from cells. Elevated levels of soluble mesothelin in ~50% of patients with mesothelioma and ovarian cancer but have not been observed in pancreatic adenocarcinoma.
- Normal expression on mesothelial cells lining pleura, peritoneum, and pericardium.
- Biological function remains unclear but has been shown to bind to CA-125 (MUC16) and may have a role in cellular adhesion, tumor invasion and metastasis.

## Expression by malignant tissues

- Pancreatic adenocarcinoma (100%)
- Epithelial mesothelioma (100%)
- Ovarian carcinoma (85-100%)
- Lung adenocarcinoma (50%)




**Pastan and Hassan *Cancer Res* 2014; Chang and Pastan *PNAS* 1996; Carpenito et al, *PNAS* 2009**

# Penn Clinical Trials using SS1 and M5 Mesothelin-targeting CAR T cells for the treatment of solid tumors

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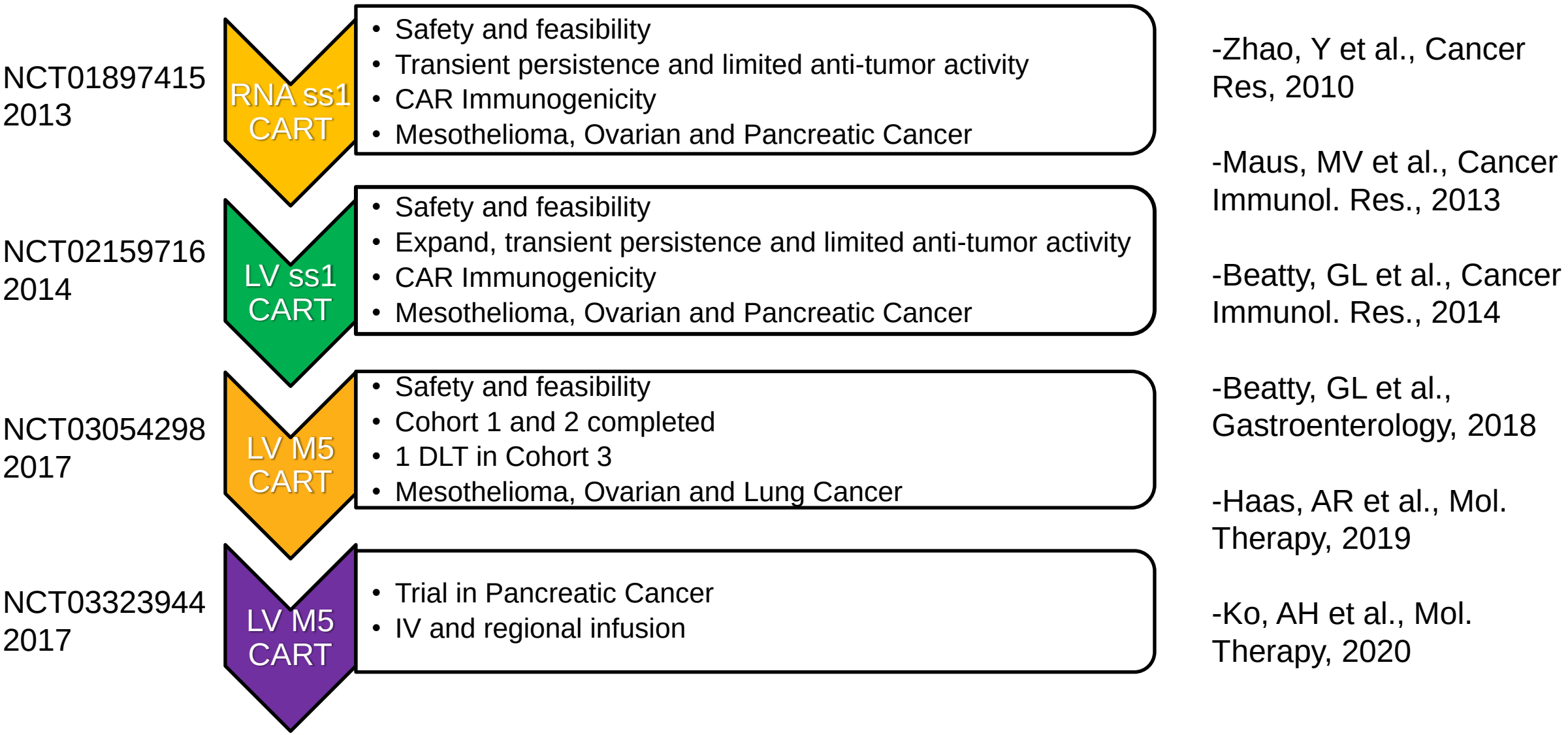


-Zhao, Y et al., Cancer Res, 2010

-Maus, MV et al., Cancer Immunol. Res., 2013

-Beatty, GL et al., Cancer Immunol. Res., 2014

-Beatty, GL et al., Gastroenterology, 2018

-Haas, AR et al., Mol. Therapy, 2019

-Ko, AH et al., Mol. Therapy, 2020

# Activity of SS1 Mesothelin RNA CAR in Metastatic PDA: Clinical Responses

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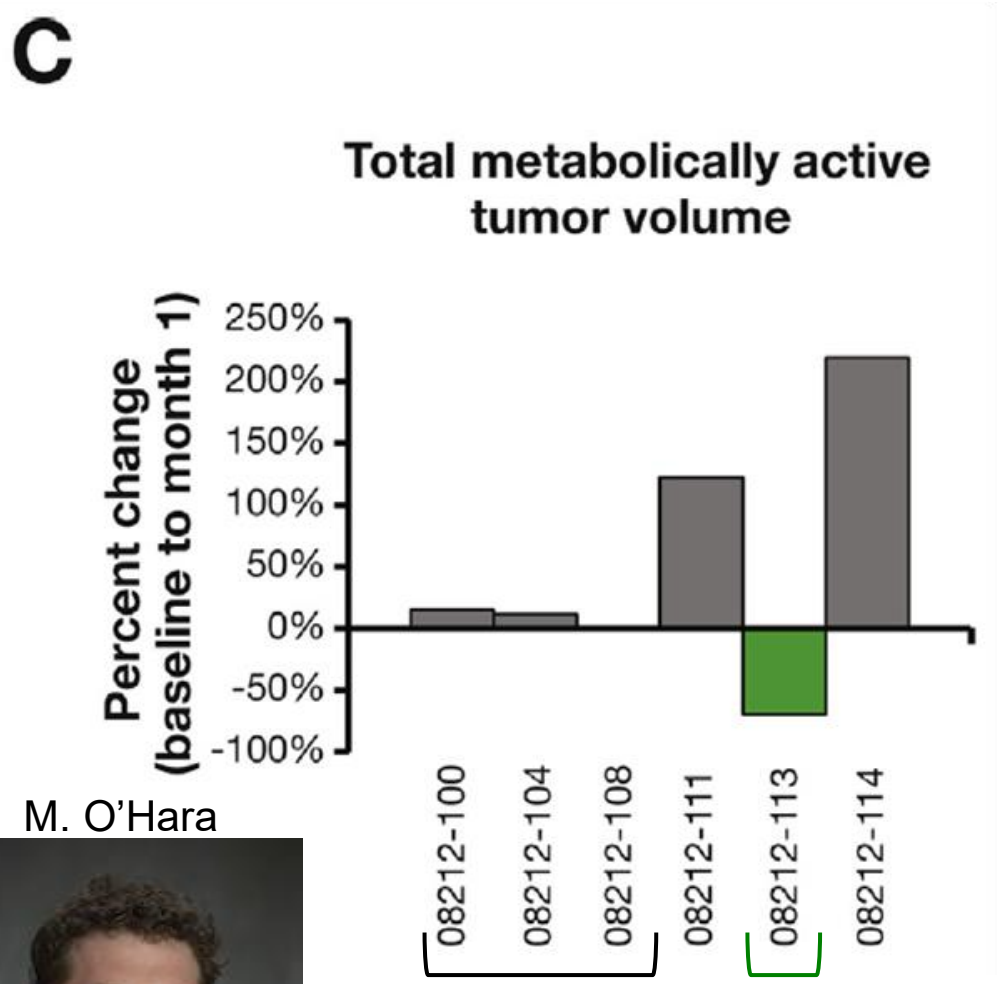
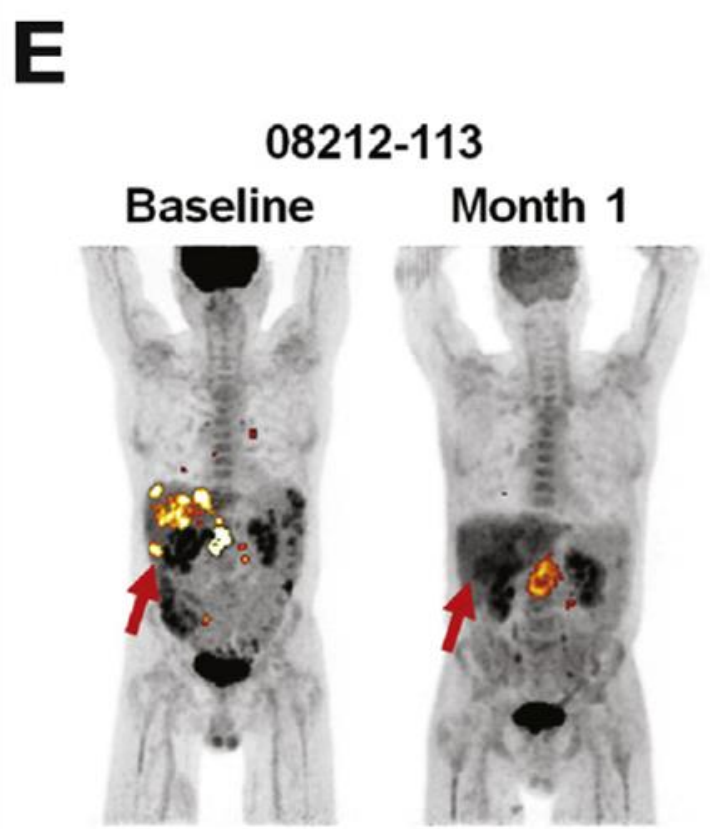
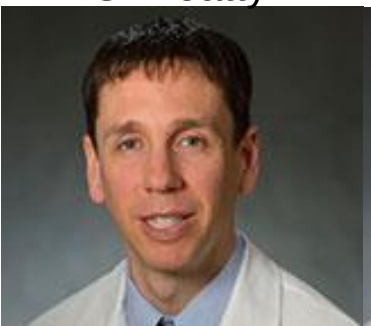
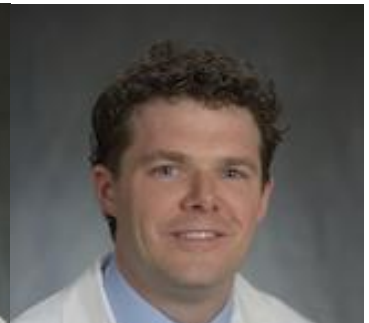



G. Beatty



M. O'Hara



Beatty et al. Gastroenterology, 2018

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# Summary: Mesothelin CAR T Cells

- **CAR T targeting mesothelin have antitumor activity in mesothelioma and pancreatic cancer**
- **The optimal schedule and route of administration of CAR T cells remains to be defined**
- **CAR T cell trafficking to tumor and evidence of spreading immunity**
- **Checkpoint blockade with PD-1 and CTLA-4 antagonists augments the activity of CAR T cells in preclinical models and early stage clinical trials**

1. Beatty GL, et al . Mesothelin-specific chimeric antigen receptor mRNA-engineered T cells induce anti-tumor activity in solid malignancies. *Cancer Immunol Res*. 2014;2:112.
2. Beatty GL, et al. Activity of Mesothelin-specific Chimeric Antigen Receptor T cells Against Pancreatic Carcinoma Metastases in a Phase 1 Trial. *Gastroenterology*. 2018; pii: S0016-5085(18)30323.

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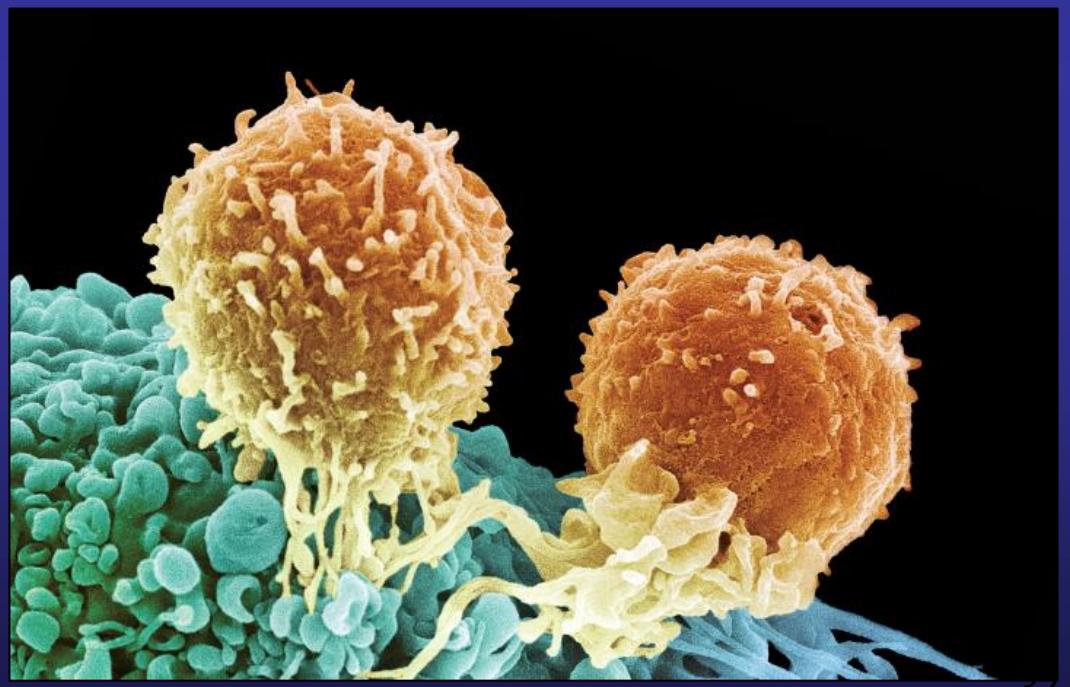
# Multiplex Engineered T Cells in Cancer



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# NY-ESO-1–specific TCR–engineered T cells mediate sustained antigen-specific antitumor effects in myeloma

Aaron P Rapoport[1,8], Edward A Stadtmauer[2,8], Gwendolyn K Binder-Scholl[3,8], Olga Goloubeva[1,4], Dan T Vogl[2], Simon F Lacey[2,5], Ashraf Z Badros[1], Alfred Garfall[2], Brendan Weiss[2], Jeffrey Finklestein[4,5], Irina Kulikovskaya[2,5], Sanjoy K Sinha[6], Shari Kronsberg[1,4], Minnal Gupta[2,5], Sarah Bond[7], Luca Melchiori[3], Joanna E Brewer[3], Alan D Bennett[3], Andrew B Gerry[3], Nicholas J Pumphrey[3], Daniel Williams[3], Helen K Tayton- Martin[3], Lilliam Ribeiro[3], Tom Holdich[3], Saul Yanovich[1], Nancy Hardy[1], Jean Yared[1], Naseem Kerr[5], Sunita Philip[1], Sandra Westphal[1], Don L Siegel[2,5], Bruce L Levine[2,5], Bent K Jakobsen[3], Michael Kalos[2,5,8] & Carl H June[2,5]

Encouraging clinical responses were observed in 16 of 20 patients (80%) with advanced disease, with a median progression-free survival of 19.1 months. NY-ESO-1–LAGE-1 TCR–engineered T cells were safe, trafficked to marrow and showed extended persistence that correlated with clinical activity against antigen-positive myeloma.

VOLUME 21 | NUMBER 8 | AUGUST 2015 NATURE MEDICINE

- **Relapse associated with loss of gene modified cells**
- **Evidence suggests that NY-ESO-1 T cells become exhausted**
- **How to enhance survival and function?**

# Hypothesis and Pre-clinical Work

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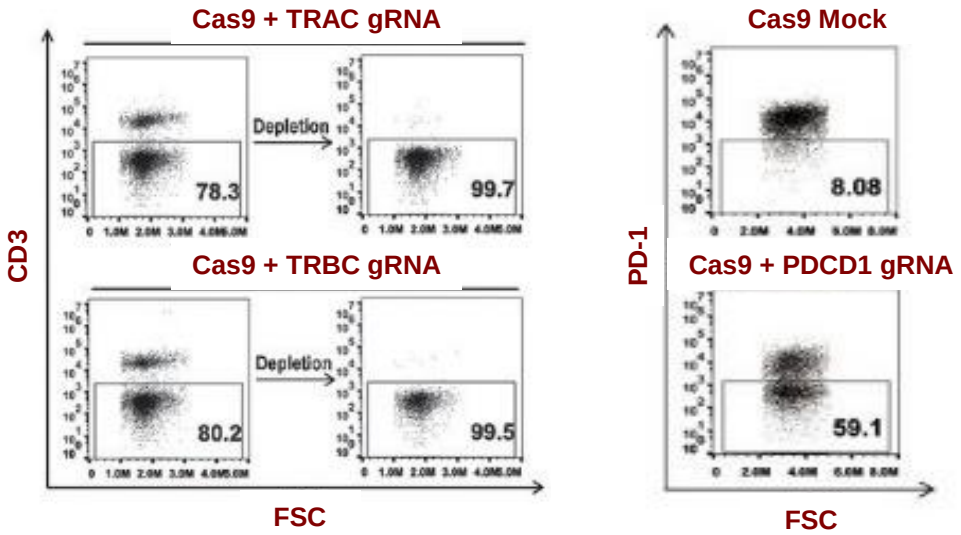
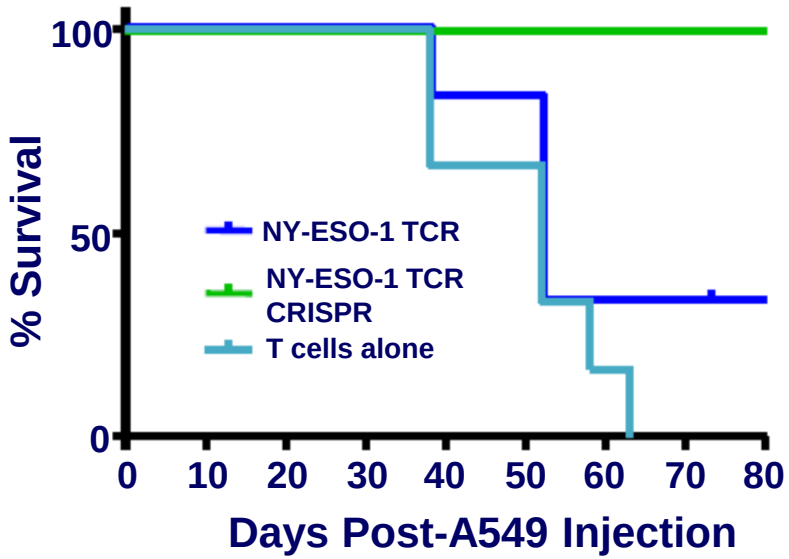
- **Hypothesis:**
  - **Removal of genes encoding the endogenous TCR, TCRα (*TRAC*) and TCRβ (*TRBC*), would enhance NY-ESO-1 TCR activity and reduce autoimmunity**
  - **Removal of PD-1 (*PDCD1*) would enhance activity and persistence**

- **We previously demonstrated CRISPR/Cas9 and *TRAC*, *TRBC* and *PDCD1* targeting gRNAs could be successfully introduced via electroporation in preclinical models to disrupt gene expression**

## CRISPR/Cas9 Editing of T cells




## Overall Survival




Ren et al, *Clin. Cancer Res.* 2016

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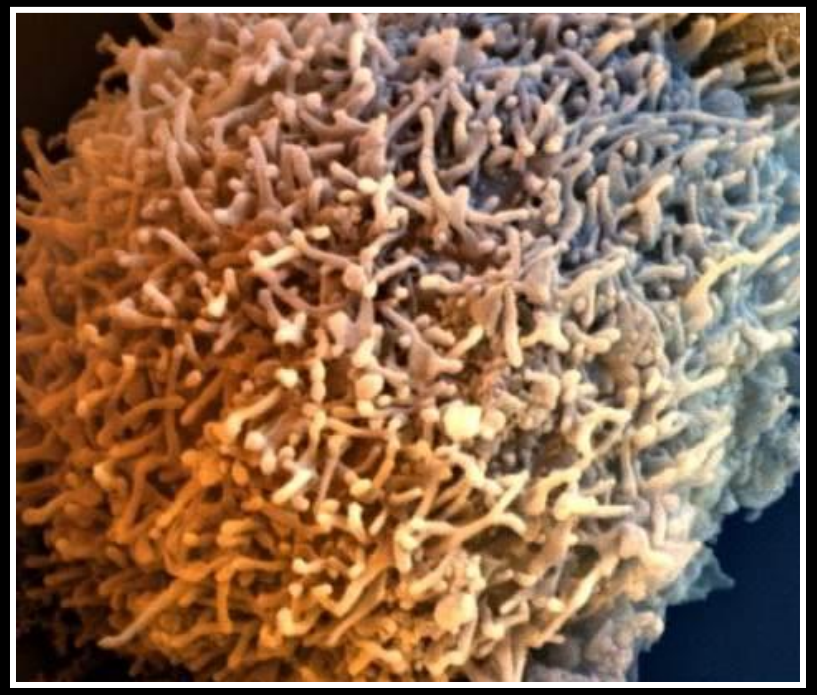
# The Era of Synthetic Biology
# Advanced Cellular Re-Programming



- Logic Gated Boolean CAR's- "And", "Or", Not
- Safety Switches
- Conditional/Stealth CAR's
- Armored CAR's

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# NY-ESO-1 TCR CRISPR Triple Edited (TCRα TCRβ PD1) T Cell

## Study Objectives

| | |
|---|---|
| Objectives | Primary: Determine safety profile of a single infusion of autologous t cells modified to express NY-ESO-1 transgenic TCR and gene edited at the endogenous TCR and PD-1 (CRISPR edited T cells)<br><br>Secondary:<br>1. Describe anti-tumor responses and survival after infusion<br>2. Evaluate manufacturing feasibility<br>3. Determine engraftment, persistence, and trafficking of NY-ESO-1 redirected CRISPR cells<br>4. Evaluate bioactivity of NY-ESO-1 redirected CRISPR cells<br>5. Describe the incidence of immunogenicity |

Adult patients HLA-A2*0201 positive who have relapsed/refractory tumors expressing NY-ESO-1 antigen. Patients with myeloma, synovial sarcoma, melanoma

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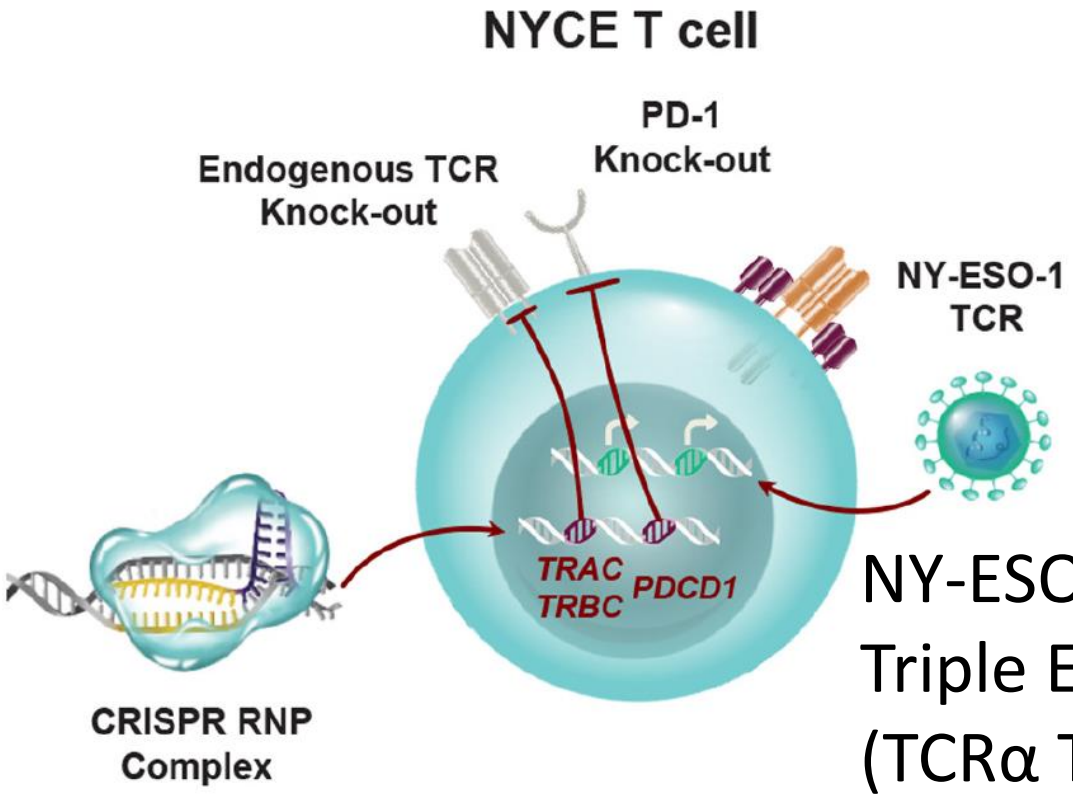
# CRISPR-engineered T cells in patients with refractory cancer

Edward A. Stadtmauer,[1,2*†] Joseph A. Fraietta,[2,3,4,5*] Megan M. Davis,[5,6] Adam D. Cohen,[1,2] Kristy L. Weber,[2,7] Eric Lancaster,[8] Patricia A. Mangan,[1] Irina Kulikovskaya,[5] Minnal Gupta,[5] Fang Chen,[5] Lifeng Tian,[5] Vanessa E. Gonzalez,[5] Jun Xu,[5] In-young Jung,[4,5] J. Joseph Melenhorst,[3,5,6] Gabriela Plesa,[5] Joanne Shea,[5] Tina Matlawski,[5] Amanda Cervini,[5] Avery L. Gaymon,[5] Stephanie Desjardins,[5] Anne Lamontagne,[5] January Salas-Mckee,[5] Andrew Fesnak,[5,6] Donald L. Siegel,[5,6] Bruce L. Levine,[5,6] Julie K. Jadlowsky,[5] Regina M. Young,[5] Anne Chew,[5] Wei-Ting Hwang,[9] Elizabeth O. Hexner,[1,2] Beatriz M. Carreno,[3,5,6] Christopher L. Nobles,[4] Frederic D. Bushman,[4] Kevin R. Parker,[10] Yanyan Qi,[11] Ansuman T. Satpathy,[10,11] Howard Y. Chang,[10,12] Yangbing Zhao,[5,6] Simon F. Lacey,[5,6*] Carl H. June[2,3,5,6*†]




NY-ESO-1 TCR CRISPR
Triple Edited
(TCRα TCRβ PD1) T Cell

# Feasibility of multiplex human genome editing with CRISPR-Cas9: Cell Product

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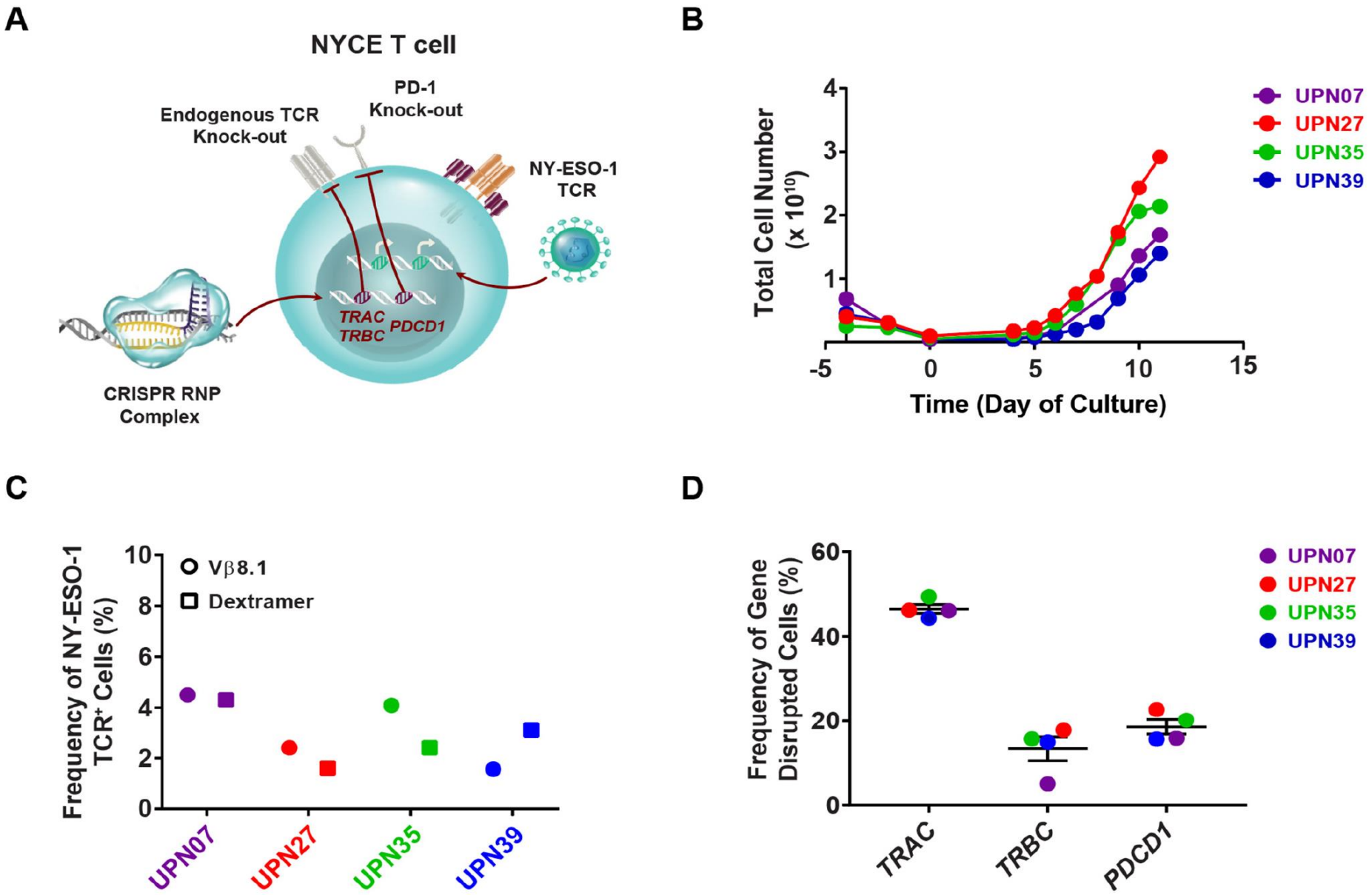


Stadtmauer et al., Science. 2020 Feb 28;367(6481):eaba7365

# Expansion, Persistence and Trafficking of NYCE T cells: *NY-ESO-1 TCR T cells (PCR)*

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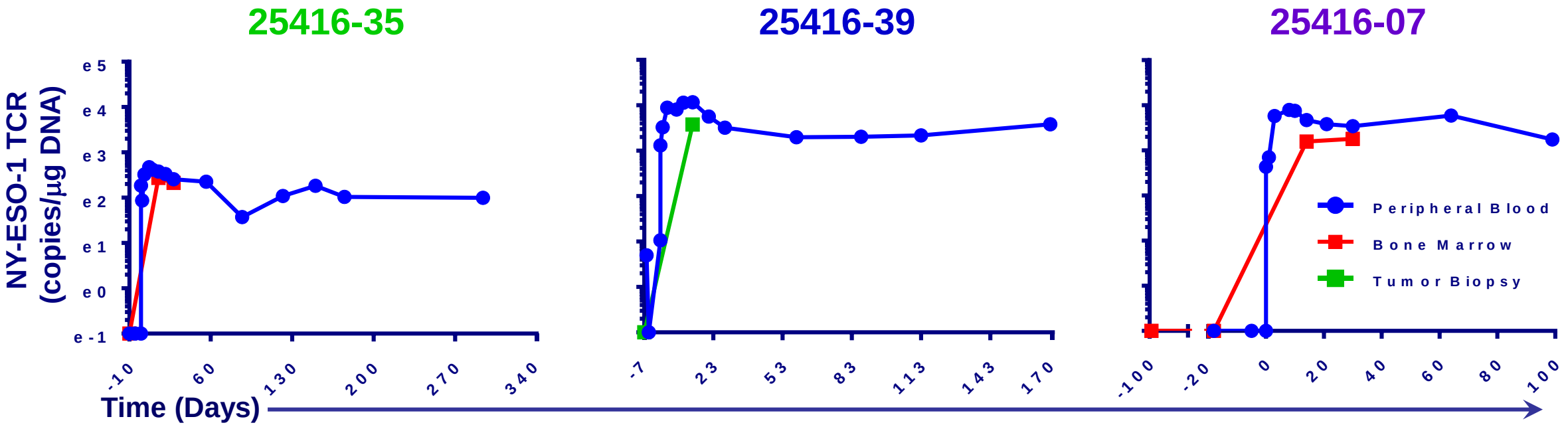


- **There is rapid expansion and stable persistence of T cells expressing the NY-ESO-1 transgenic TCR as measured by qPCR in all 3 patients**
  - **The stable PK of NY-ESO-1 expressing T cells is very different from the PK of CAR T cells which tends to decrease more quickly**
- **Clear trafficking of T cells to the tumor**
  - **The levels of T cells expressing the NY-ESO-1 TCR in bone marrow and tumor is similar to blood**

**Stadtmauer et al., Science. 2020 Feb 28;367(6481):eaba7365**

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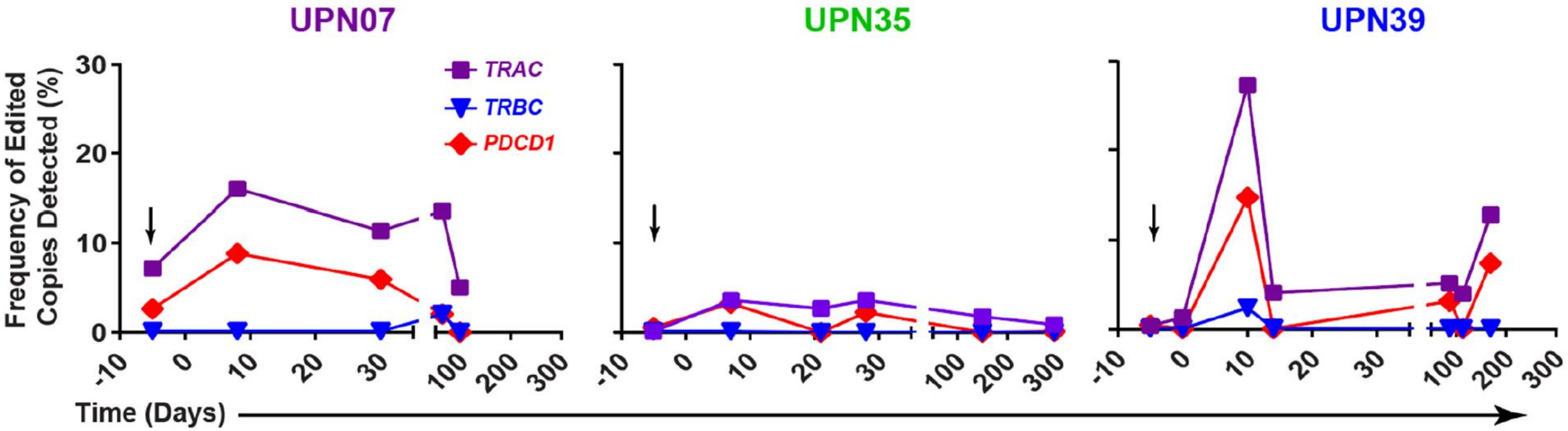
# NYCE CRISPR Genome-Edited Cells: Persistence in Blood of TRAC, TRBC, and PDCD1 edits by dPCR




- TRAC and PD1 have highest frequency
- TRBC was lowest efficiency in vitro, and also in vivo
- Pt 07 shortest follow up

**Stadtmauer et al., Science. 2020 Feb 28;367(6481):eaba7365**

# Di-genic and Tri-genic Editing at the level of a single human genome

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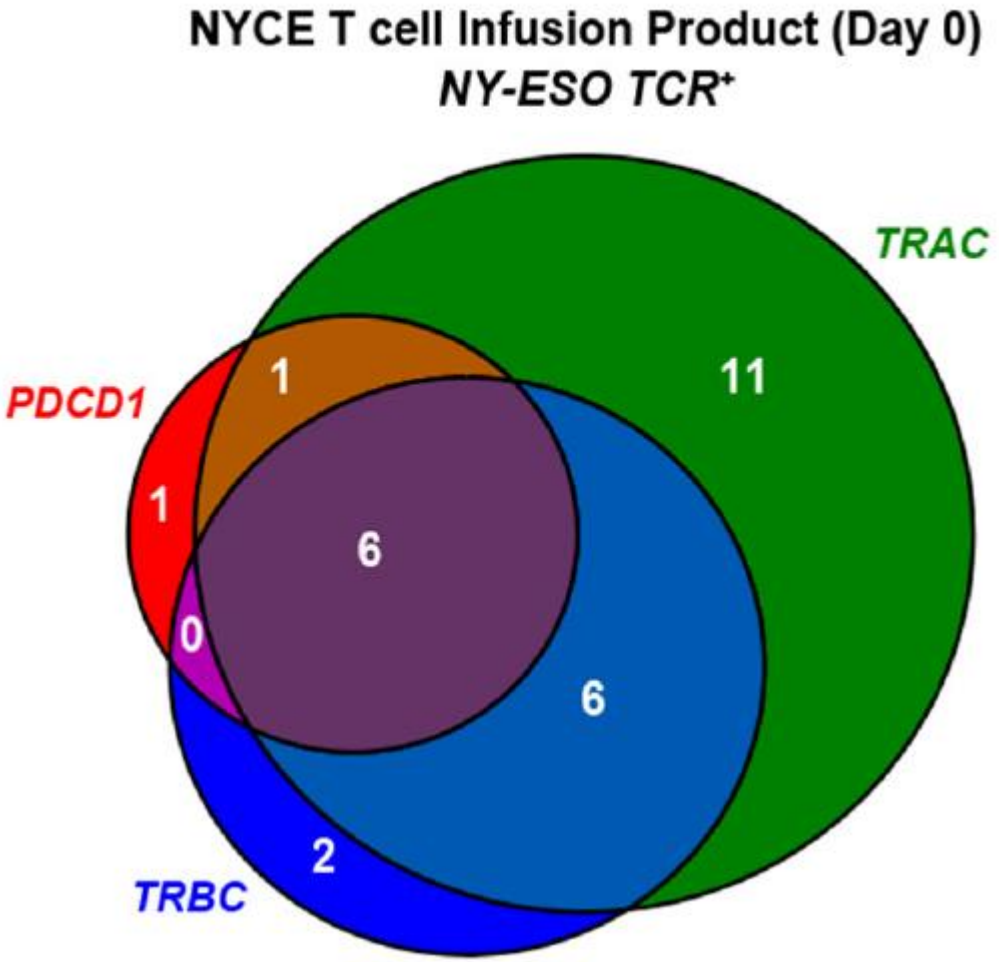


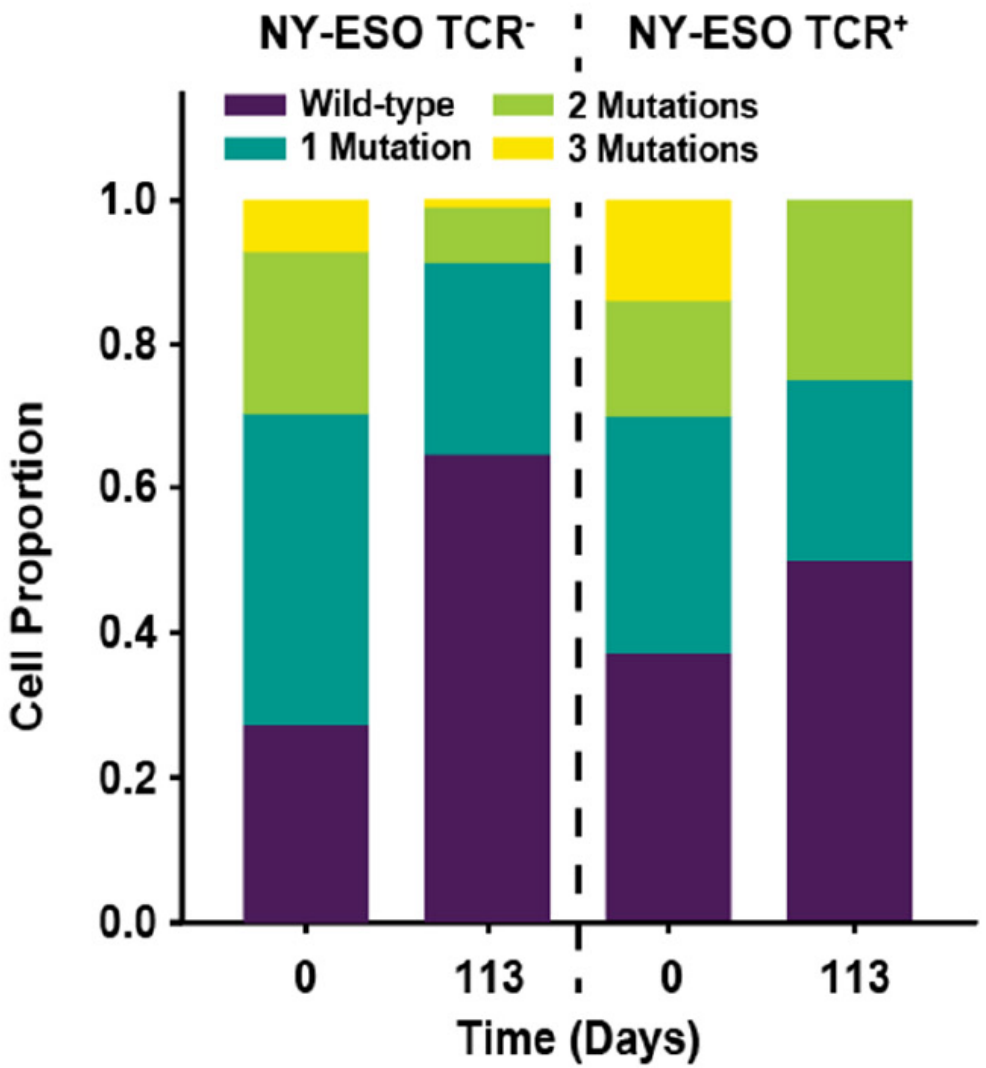


Stadtmauer et al., Science. 2020 Feb 28;367(6481):eaba7365

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# NY-ESO-1 CRISPR (TCR-PD1) Triple Edited T Cell Study Summary

1. **A single infusion of CRISPR/Cas9 triple edited T cells is safe in 3 patients**
   - **No cytokine release syndrome, no severe adverse events**
   - **Best response is stable disease**
2. **Persistent engraftment of the edited T cells for at least 6 months**
   - **Suggests immunogenicity of Cas9 is not occurring under these conditions**
   - **Studies to determine impact of T cells on NY-ESO1 and LAGE1 expression are pending**
3. **Up to 10% of the TCR-modified T cells have PD1 disruption at 1 to 6 months**
   - **No evidence of autoimmunity**
4. **This trial: 2016-17 gene editing technology. New trial: latest improved gene editing technology**


IND 17297 and Clinicaltrials.gov NCT03399448
Sponsor: Tmunity and Parker Institute for Cancer Immunotherapy

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Table S1. NEC3 CAR-T product release specifications.

| Assay | Release Specification |
|---|---|
| Cell viability on sentinel vial | >70% |
| Transduction efficiency by flow cytometry (Net Vβ8.1 surface expression) | ≥ 2% |
| Residual bead number | < 100 beads / 3e6 cells |
| Endotoxin | Negative |
| Mycoplasma | Negative |
| BACTEC culture | No growth at day 7 |
| IL-2 independent growth (long-term culture) | No proliferation in absence of IL-2 |
| Fungal culture | No growth at day 7 |
| Transduction efficiency for transgene integration (qPCR WPRE copy number) | ≥ 0.02 - ≤ 5 average copies/cell |
| RCL (VSVg copy number) | Decreasing from day 5 to post-harvest or < 50 average copies/ug DNA |
| Disruption of *TRAC* gene (TCR-alpha editing) | Detectable disruption |
| Disruption of *TRBC* gene (TCR-beta editing) | Detectable disruption |
| Disruption of *PDCD1* gene | Detectable disruption |
| Residual Cas9 protein | Decreasing Cas9 concentration from day 0 to harvest |

Stadtmauer EA, et al. Science. 2020 Feb 28;367(6481):eaba7365. doi: 10.1126/science.aba7365.

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# What a Gene Editing Investigator May Be Asked

- Worst case scenario: induced off-target effects that induce transformation, and a form of hematologic malignancy
- FDA: Off target sites identification needs to be based on not only in silico analysis but also on unbiased assays.
- # gRNAs evaluated, detailed description of manufacture, # non-complementary bases tolerated, sequence of Cas9, purity, ratio of free vs complexed protein, stability, residual
- Laboratory test, validated laboratory test, CLIA test
- Long term culture assay
- Cell product potency

Penn Medicine
Center for Cellular Immunotherapies

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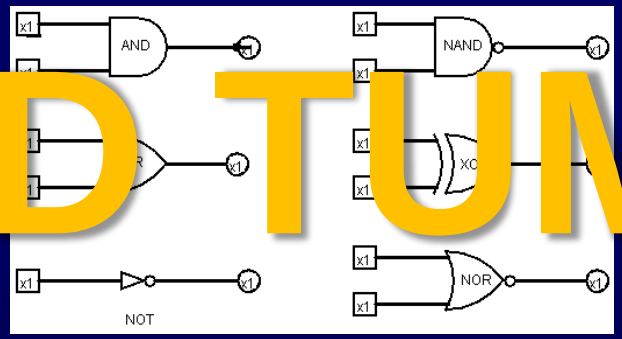
# Synthetic Biology:
# The Advanced CAR Toolbox




# SOLID TUMORS

- Logic Gated Boolean CAR's- "And", "Or", Not
- Checkpoint Resistant CAR's
- Safety Switches
- Conditional/Stealth CAR's

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# Evolution in Tools

# Evolution in Manufacturing

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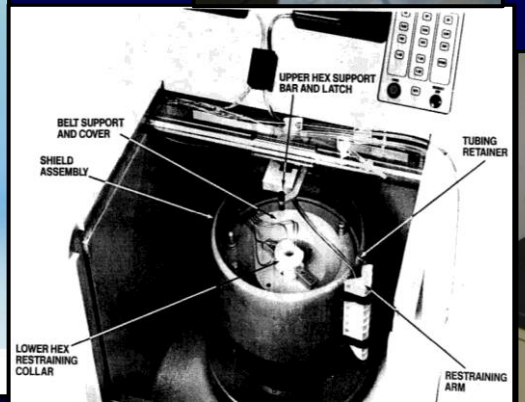
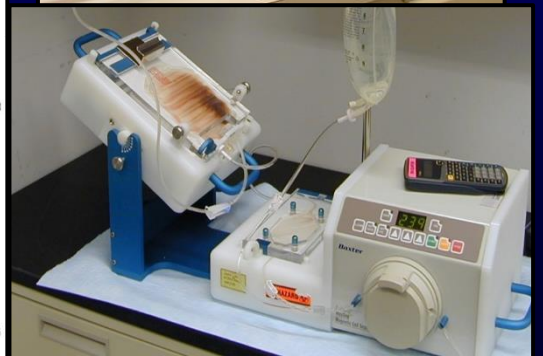
# Neolithic Cell Therapy





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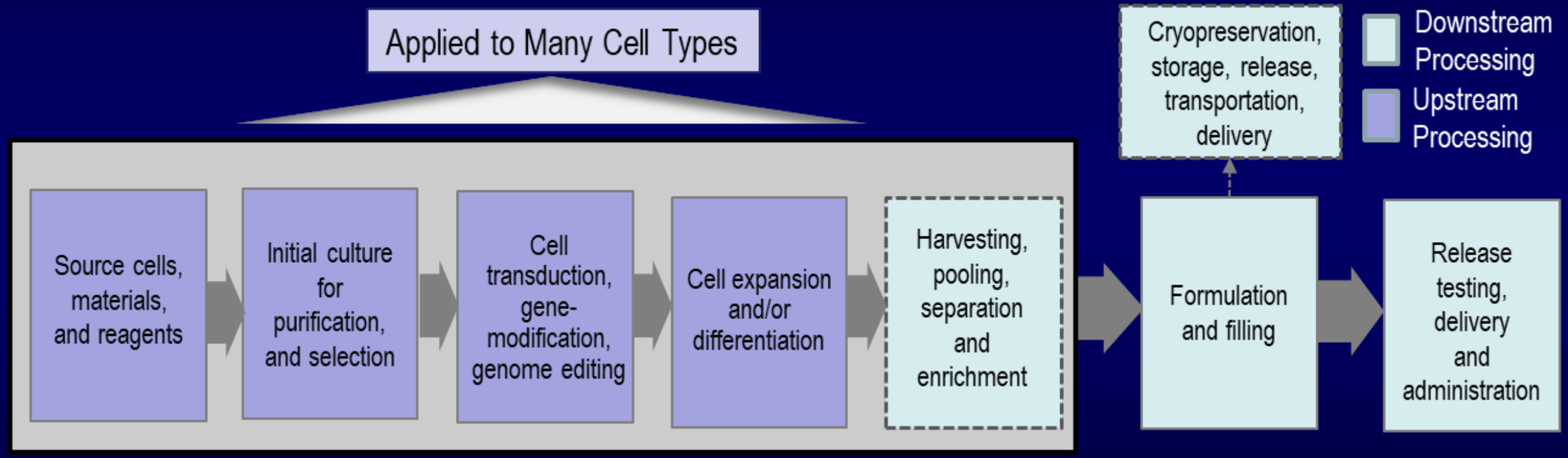
# Ex Vivo Immune Cell Engineering Processing Flow




Courtesy of Krish Roy

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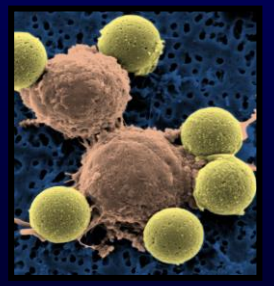
# Anthropocene Cell Therapy



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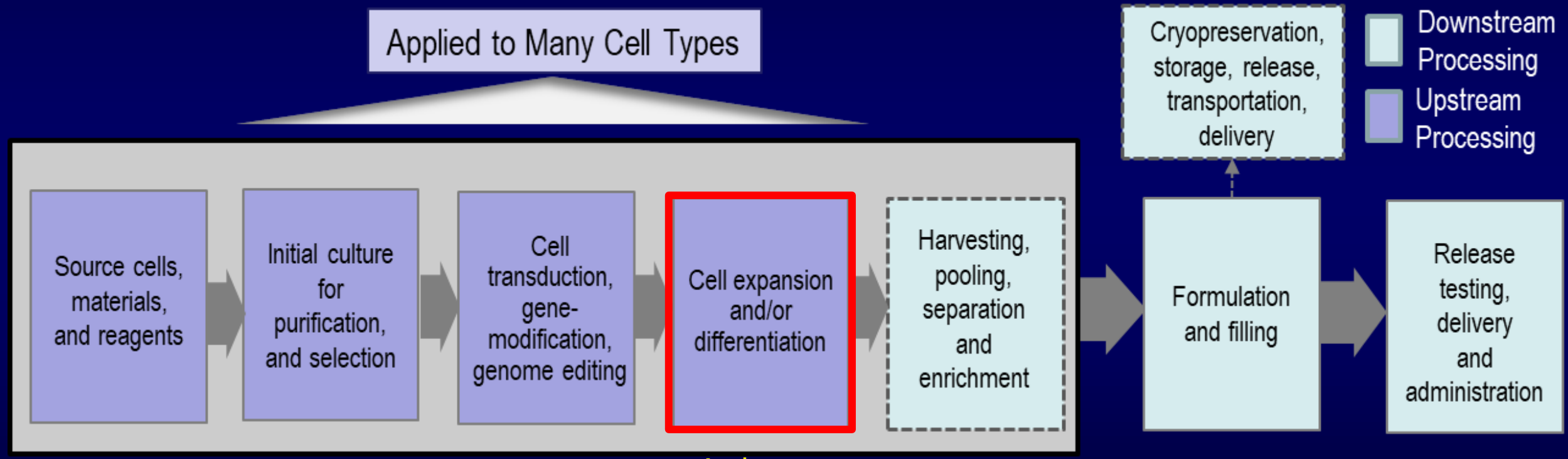
# Ex Vivo Immune Cell Engineering Processing Flow



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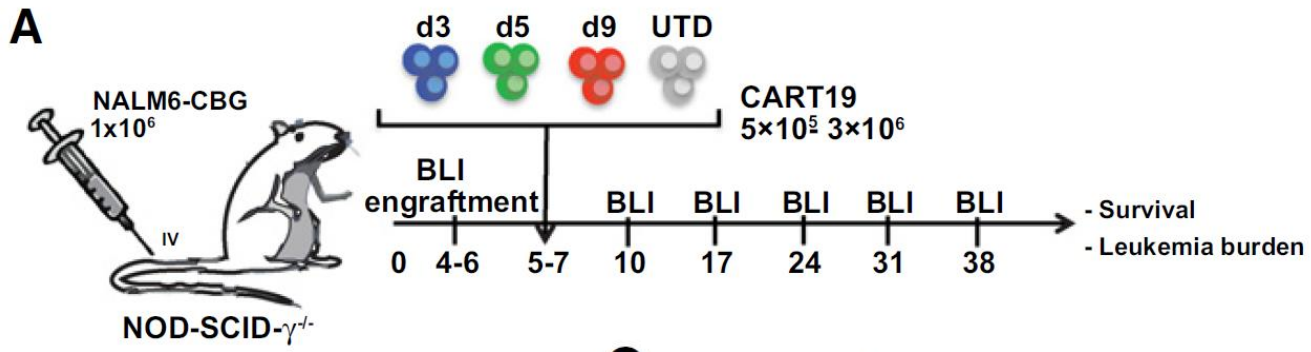
# Manufacturing Improvements: Faster CAR's

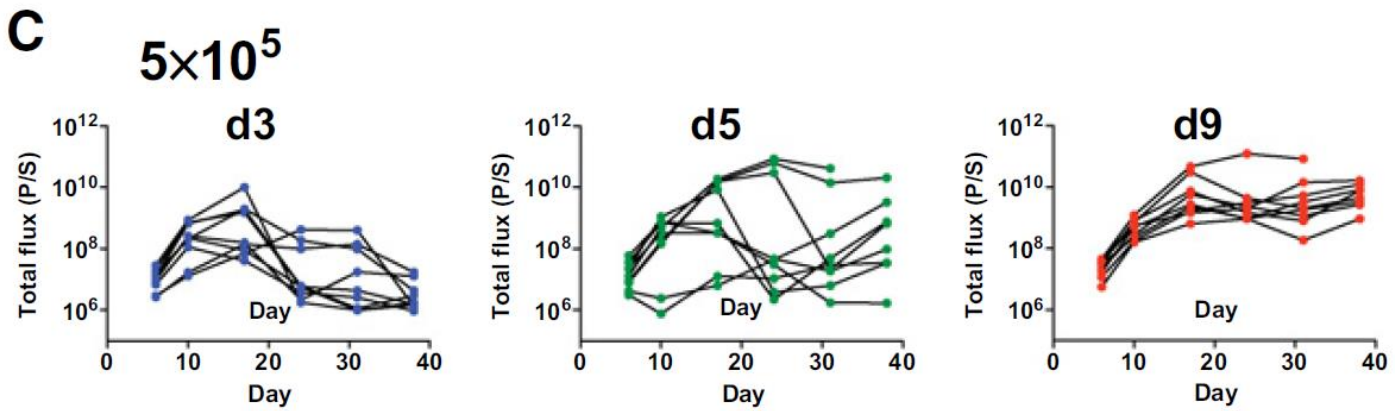
# Making Better and Cheaper CAR-T

Research Article

Cancer Immunology Research

## Reducing *Ex Vivo* Culture Improves the Antileukemic Activity of Chimeric Antigen Receptor (CAR) T Cells




Ghassemi et al, 2018

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# Successful 24-Hours Manufacture of Anti-CD19/CD22 Dual Chimeric Antigen Receptor (CAR) T Cell Therapy for B-Cell Acute Lymphoblastic Leukemia (B-ALL) – Gracell, Shanghai, China

**Table 1. Characteristics of 10 patients**

| Patient # | Gender (M/F) | Age (yrs) | BM blasts by flow cytometry (%) | Prior transplant | Prior CD19 CAR-T | CAR-T cell dose (cells/kg) | Response in 2 weeks | Response in 4 weeks | CRS | ICANS | From CAR-T to HSCT (days) | Disease status at last evaluation |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Y02001 | M | 48 | 41.6 | No | No | $6.00\times10^4$ | NR | CR MRD- | 1 | 0 | — | Relapsed with MRD+ on day116 |
| Y02002 | F | 5 | 17.7 | No | No | $6.00\times10^4$ | CR MRD- | CR MRD- | 1 | 1 | 48 | Expired from GVHD,infection on day 143 |
| Y02003 | M | 17 | 0.5 | No | Yes | $1.53\times10^5$ | CR MRD+ | — | 0 | 0 | — | Withdrew on day 14 |
| Y02004 | F | 13 | 0.1 | No | No | $1.02\times10^5$ | CR MRD- | CR MRD- | 1 | 0 | 71 | MRD-CR on day 210 |
| Y02005 | F | 11 | 24.3 | No | No | $1.50\times10^5$ | CR MRD- | CR MRD- | 1 | 0 | 57 | MRD-CR on day 187 |
| Y02006 | F | 9 | 36.6 | No | Yes | $1.00\times10^5$(second infusion $2.62\times10^5$) | CR MRD+ | CR MRD+ | 0 | 0 | — | Second infusion NR |
| Y02007 | M | 3 | 0.5 | No | No | $1.00\times10^5$ | CR MRD- | CR MRD- | 1 | 0 | 63 | MRD-CR on day 106 |
| Y02008 | F | 13 | 34.2 | Yes | Yes | $1.50\times10^5$ | CR MRD- | CR MRD- | 0 | 0 | — | Relapsed on day 84 |
| Y02009 | M | 5 | 0.1 | No | No | $1.51\times10^5$ | CR MRD- | CR MRD- | 0 | 0 | 64 | MRD-CR on day 91 |
| Y02010 | M | 12 | 63.5 | No | No | $2.25\times10^5$ | CR MRD- | CR MRD- | 2 | 0 | — | MRD-CR on day 28 |

**BM:** bone marrow, **HSCT:** hematopoietic stem cell transplantation, **CRS:** cytokine release syndrome, **ICANS:** immune effector cell-associated neurotoxicity syndrome, **NR:** no response, **MRD:** minimal residual disease, **CR:** complete remission, **MRD-CR:** MRD-negative CR, **GVHD:** graft-versus-host disease

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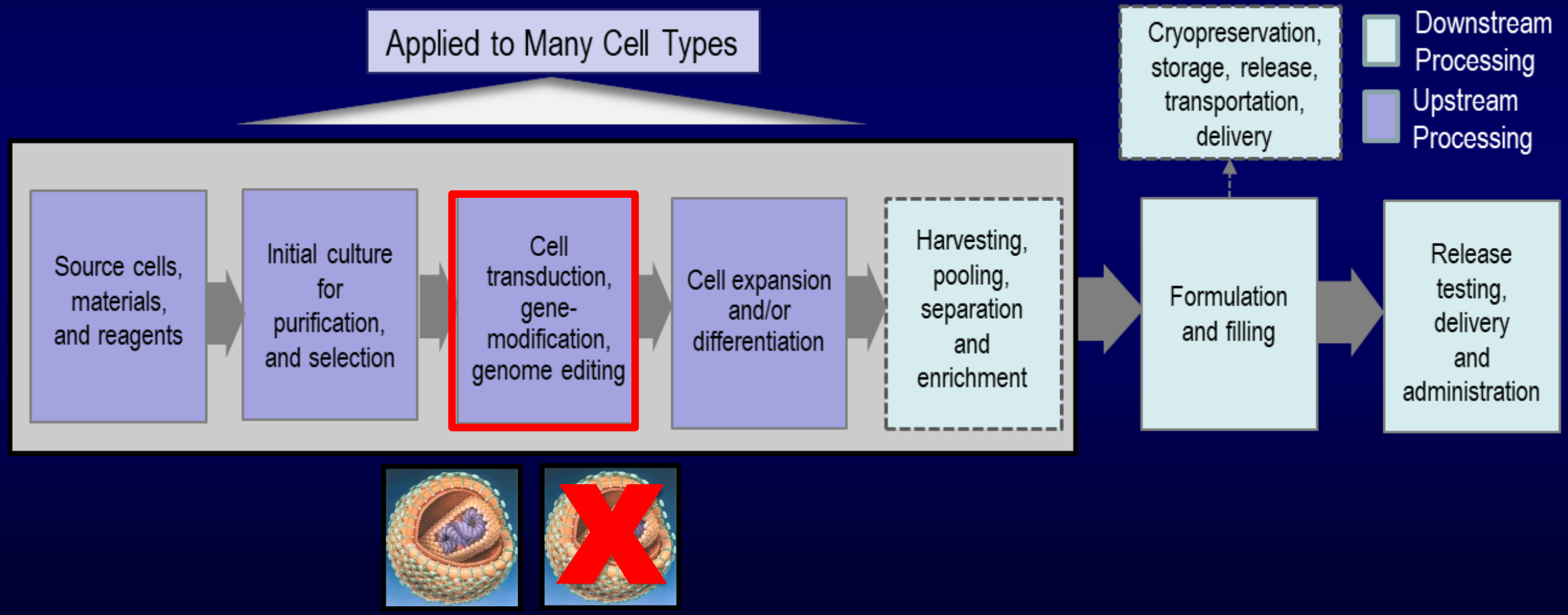
# Activated T Cell Ex Vivo Processing Flow



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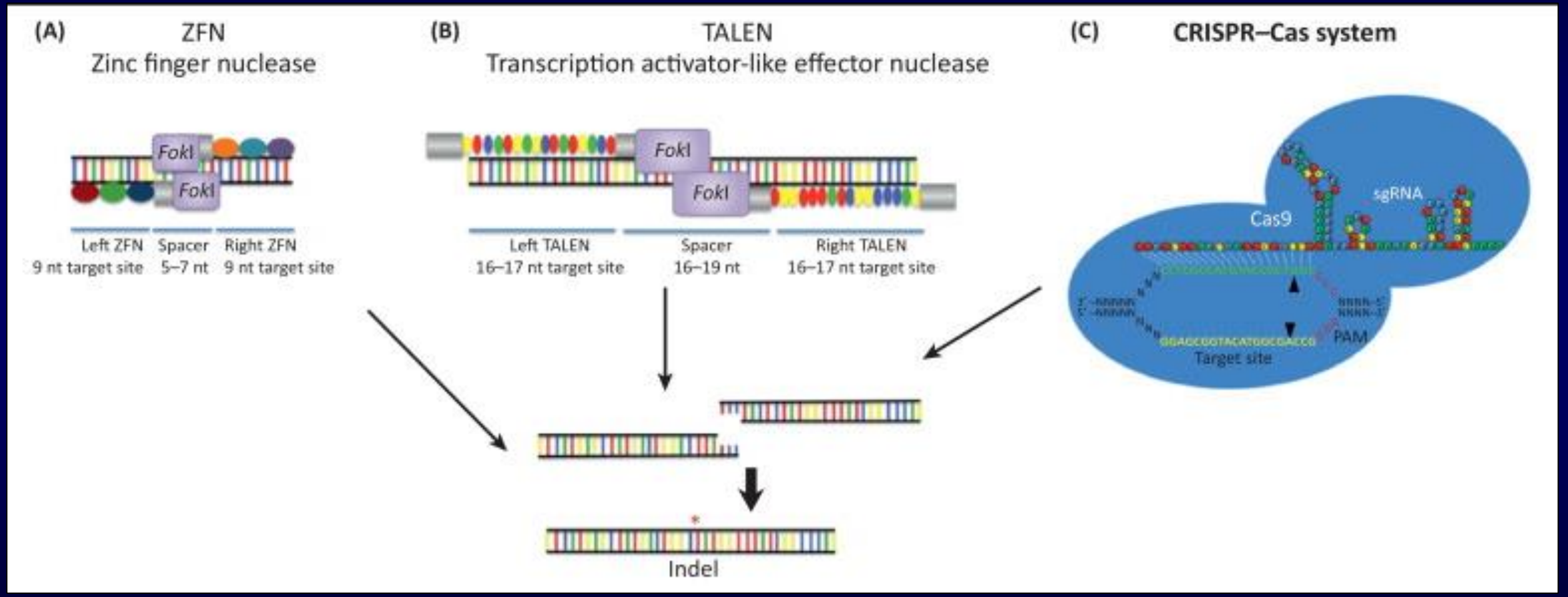
# Genomic disruption (and insertion) as therapeutic cell engineering tools




*Trends in Neurosciences* 2014 37, 106-123DOI: (10.1016/j.tins.2013.11.001)

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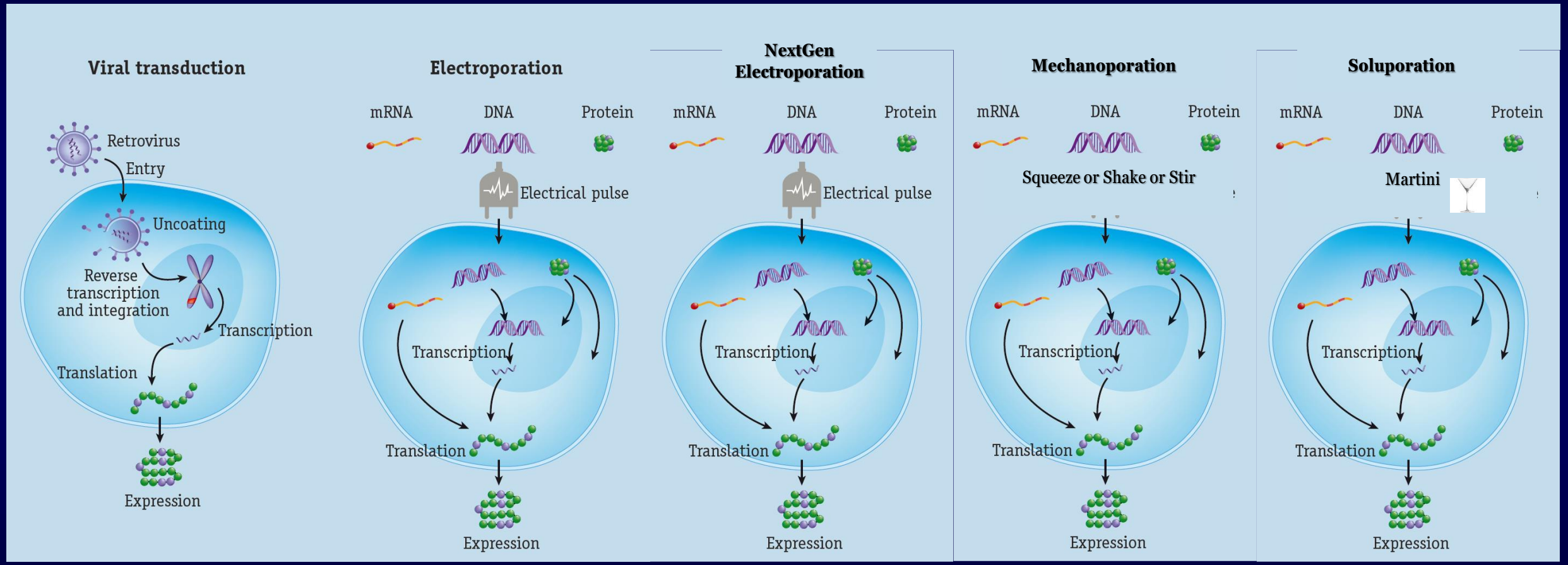
# Cellular Door Dash: Gene/Cargo Delivery



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# (Some) Critical Path Issues for Wider Patient Access

- Enhancing potency, especially for solid tumors
  - Armor, switches, combinations (CAR's)
- Manufacturing complexity: cell-based living drugs, viral vectors
  - Automation/shortened ex vivo culture
- Education & Training at all levels
  - ISCT (isctglobal.org), et al.

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- TARGETED ORGAN

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# The End of the FDA Enforcement Discretion Period and its Implications

International Society
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Thursday, June 17th
10:30–12:00 PDT / 12:30-14:00 CDT
13:30–15:00 EDT / 19:30-21:00 CEST

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SESSION CHAIRED BY

**Laertis Ikonomou, PhD**
Chair
ISCT Presidential Task Force on the Use of Unproven and/or Unethical Cell and Gene Therapy
United States

Speakers

**Wilson Bryan, MD**
Director
Office of Tissues and Advanced Therapies
FDA
United States

**Virginia Lyons, PhD**
Senator
Vermont State Senate
United States

**Leigh Turner, PhD**
Associate Professor
Center for Bioethics
University of Minnesota
United States

**Michael Lehmicke**
Director
Alliance for Regenerative Medicine
United States

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# (Some) Critical Path Issues for Wider Patient Access

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  - ISCT (isctglobal.org), et al.
- Financial complexity/affordability
  - Predictive biomarkers
  - Value based payment links price to outcome

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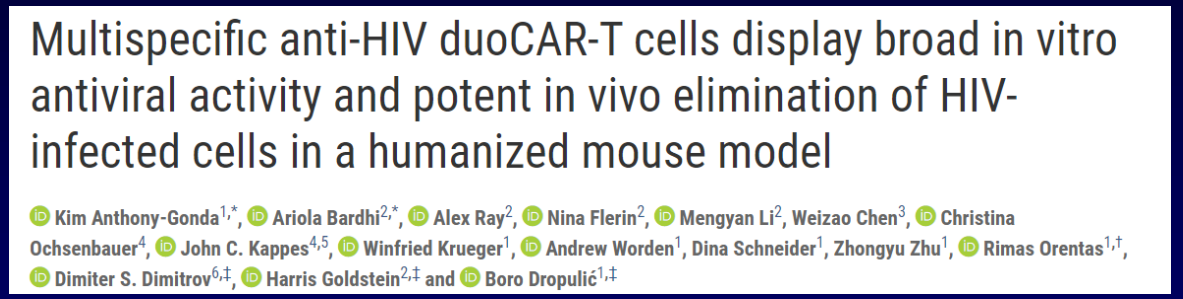
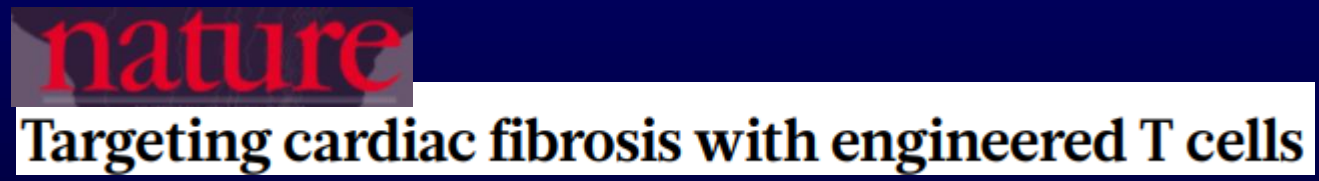
# CAR Cells Move Beyond T Cells in Oncology

- CAR T Cells for HIV/AIDS
- CAR T Cells for Autoimmunity and Organ Transplantation
- CAR Macrophages for Cancer
- CAR T Cells for Heart Failure and Fibrosis
- CAR T Cell for Aging?


Multispecific anti-HIV duoCAR-T cells display broad in vitro antiviral activity and potent in vivo elimination of HIV-infected cells in a humanized mouse model

Kim Anthony-Gonda[1,*], Ariola Bardhi[2,*], Alex Ray[2], Nina Flerin[2], Mengyan Li[2], Weizao Chen[3], Christina Ochsenbauer[4], John C. Kappes[4,5], Winfried Krueger[1], Andrew Worden[1], Dina Schneider[1], Zhongyu Zhu[1], Rimas Orentas[1,†], Dimiter S. Dimitrov[6,‡], Harris Goldstein[2,‡] and Boro Dropulić[1,‡]

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# ACKNOWLEDGEMENTS

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Abramson Cancer Center

Study Participants

FDA U.S. FOOD & DRUG ADMINISTRATION

The Leukemia & Lymphoma Society
Fighting Blood Cancers

ACGT National grants for cell and gene therapy cancer research
ALLIANCE FOR CANCER GENE THERAPY

PARKER INSTITUTE for CANCER IMMUNOTHERAPY

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# It Takes A Village