z-Movi® - 세포 간 결합과 상호작용을 이해하는 핵심, Cell Avidity

26JUN2026 김상아 / 라이노바이오 학술영업부

Rhino Bio, Inc.

LUMICKS

About LUMICKS..

Expanding into
Cell Avidity

z-Movi

Avidigo
services

2014 — 2018 — 2020 — 2024

LUMICKS founded:
introducing C-trap optical
tweezers

- Founded 2014
- 180 people, 30 nationalities

HQ Amsterdam
Boston Office
Beijing Office

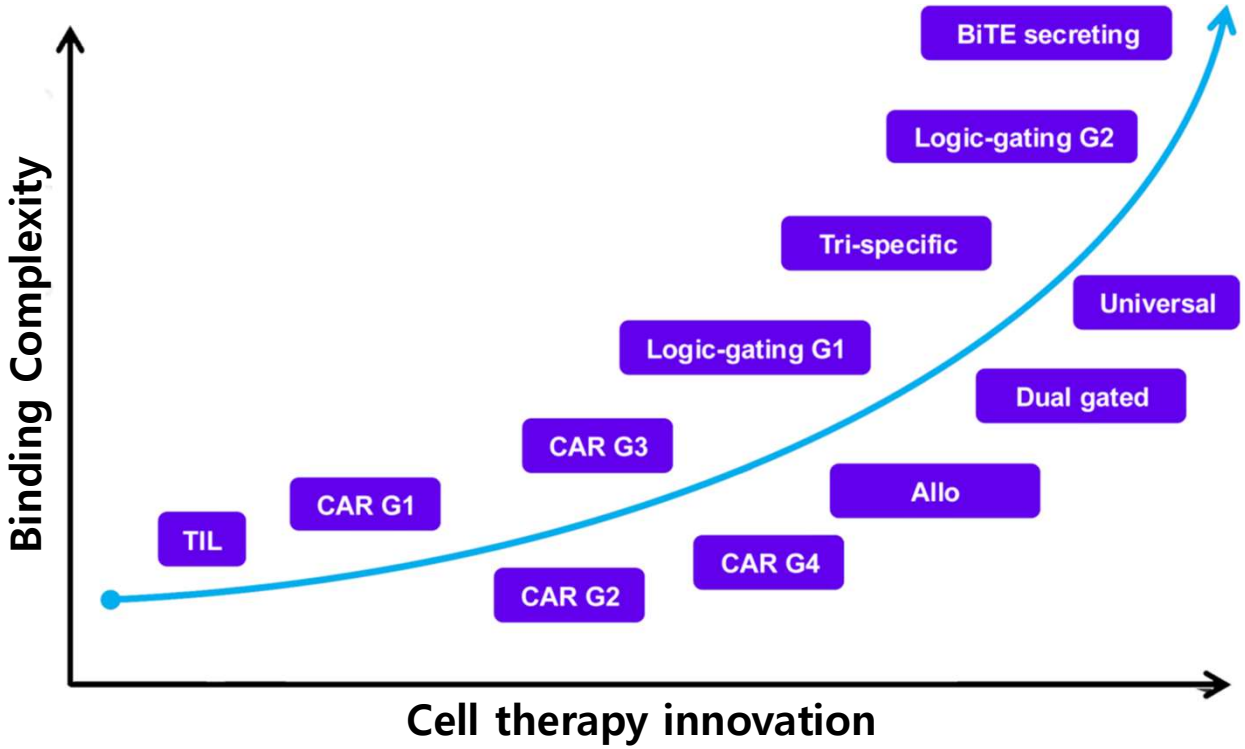
Keeping up with the complexity of binding mechanisms

Current Challenge

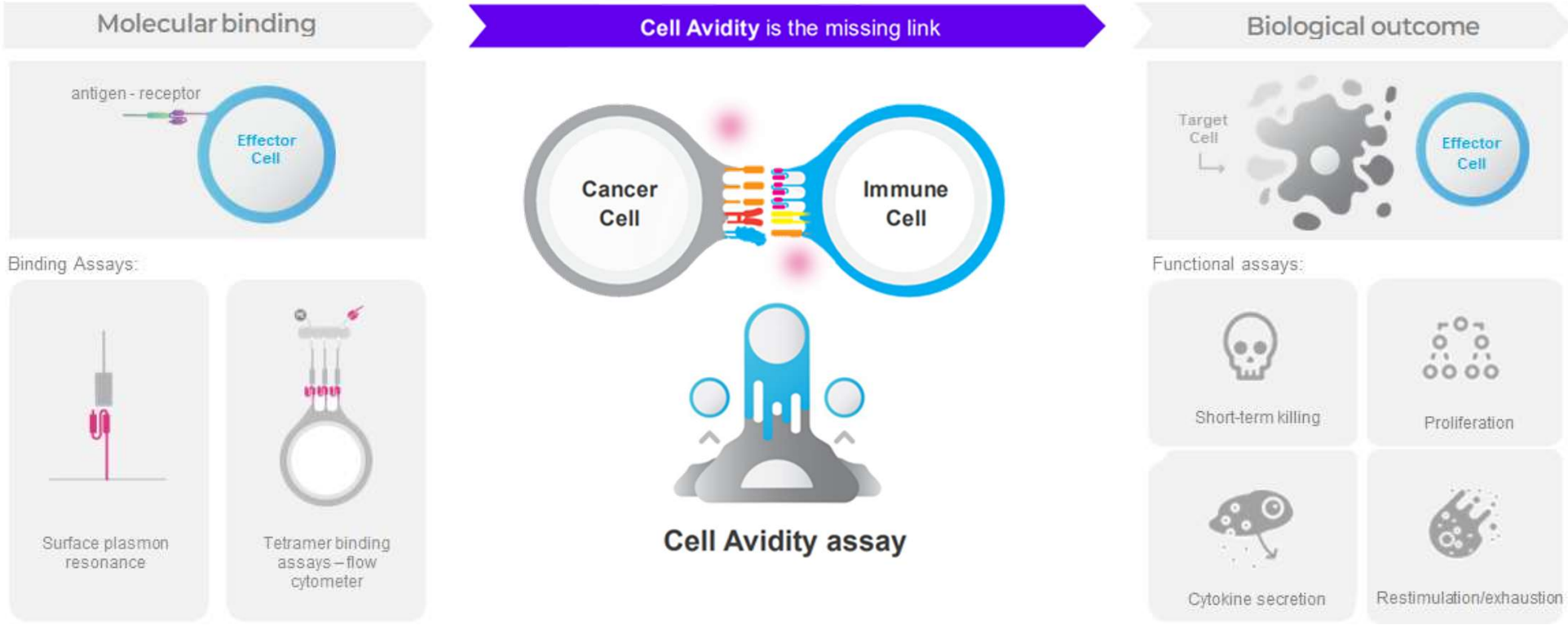
- Immunotherapies increasingly integrate multiple signaling mechanisms and engage multiple targets in parallel
- Binding mechanisms between binder and target are becoming more complex

Limitations of Molecular Binding Assays

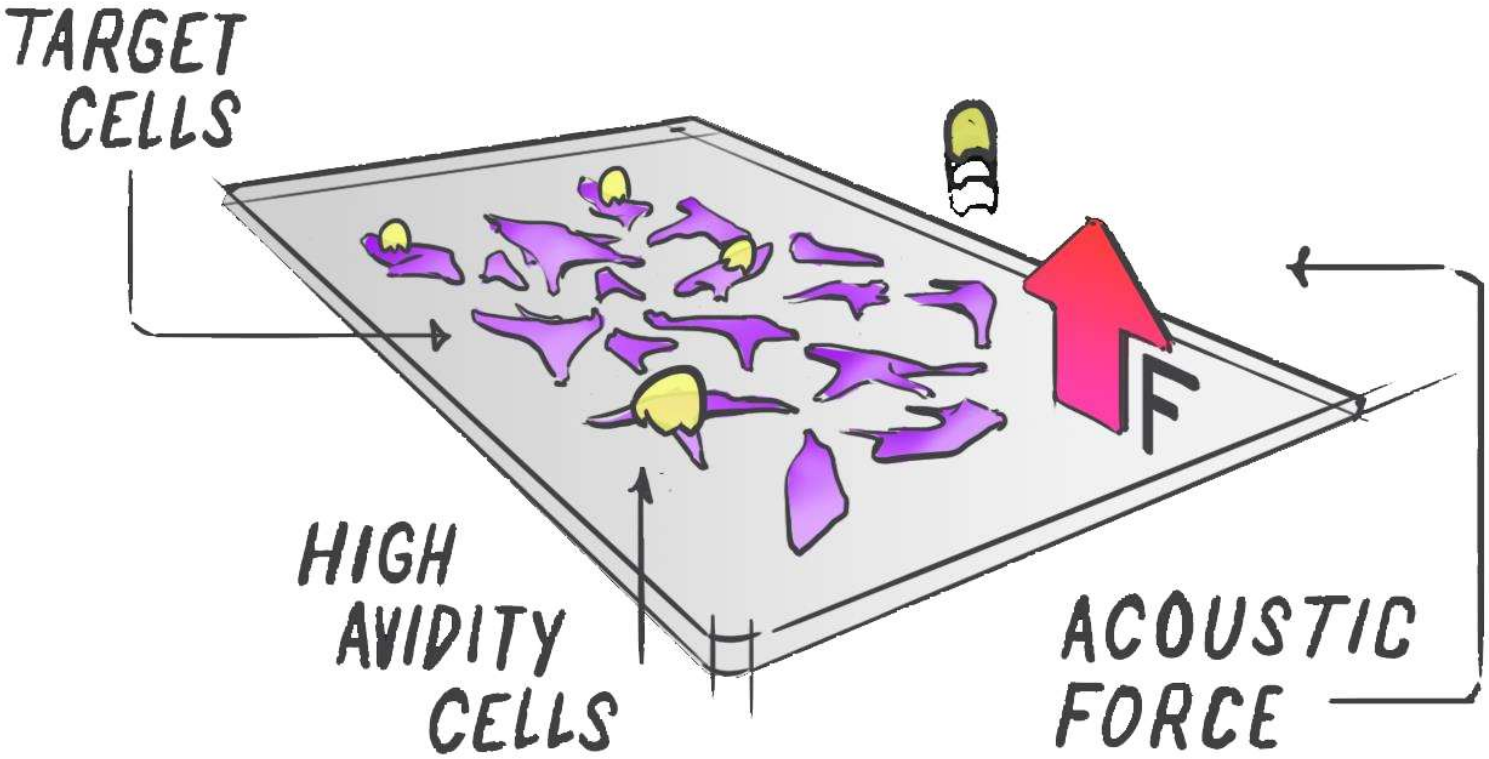
- Tetramer binding and SPR measure preconditions for binding
- Limited insights into actual cell binding
- Focus on isolated ligand–receptor interactions or molecular abundance
- May miss the cellular context
- Often do not correlate well with functional outcomes

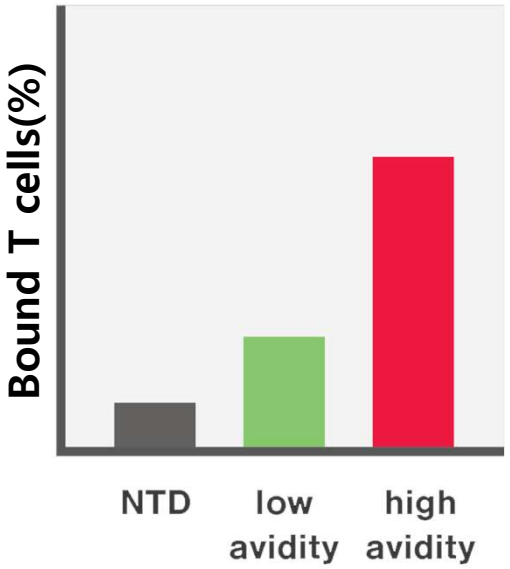


Cell Avidity: Bridging the Gap in Cell Therapy Characterization



Measuring T Cell–Tumor Cell Avidity

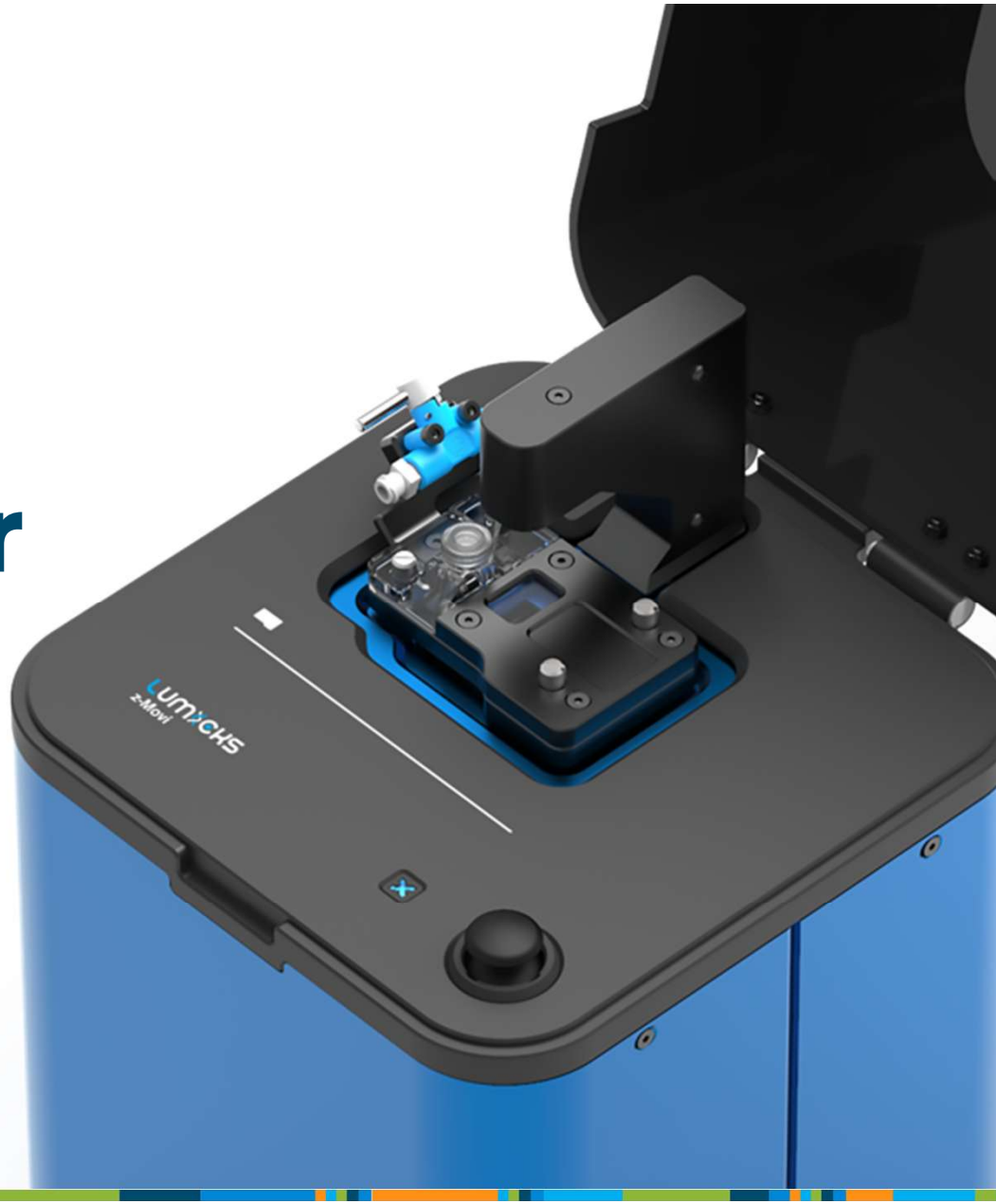




We measure the **bound T cell fraction after the force is applied** across thousands of cell-pairs, providing a **direct quantification of cell binding avidity**

z-Movi®
Cell Avidity Analyzer

The only simple, direct solution to measure **cell avidity**



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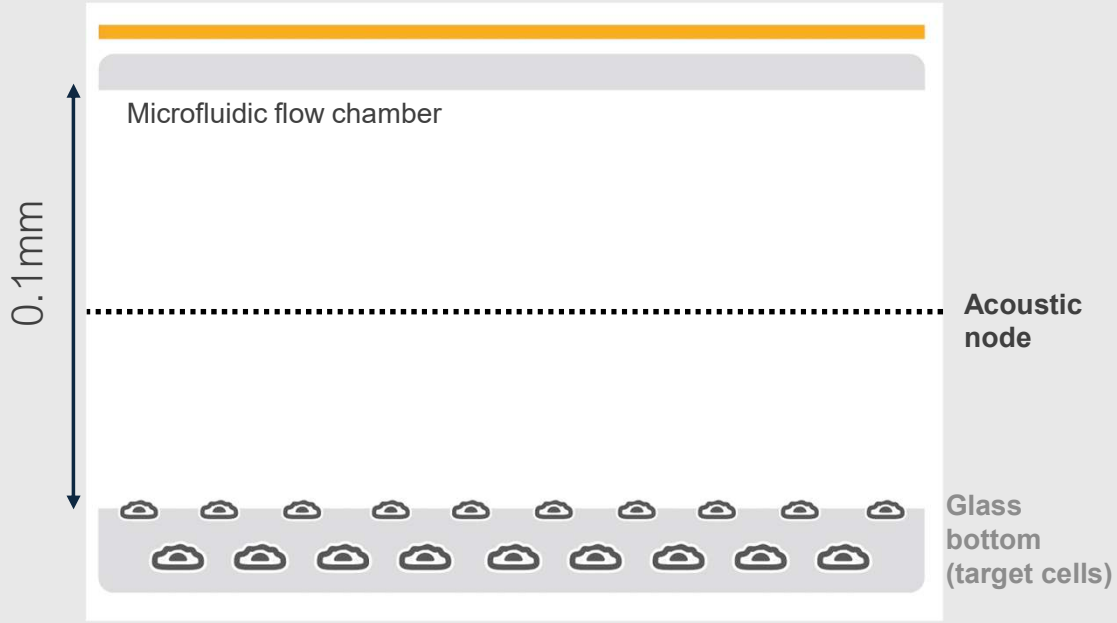
z-Movi: Benchtop Solution for Cell Avidity

Benchtop solution

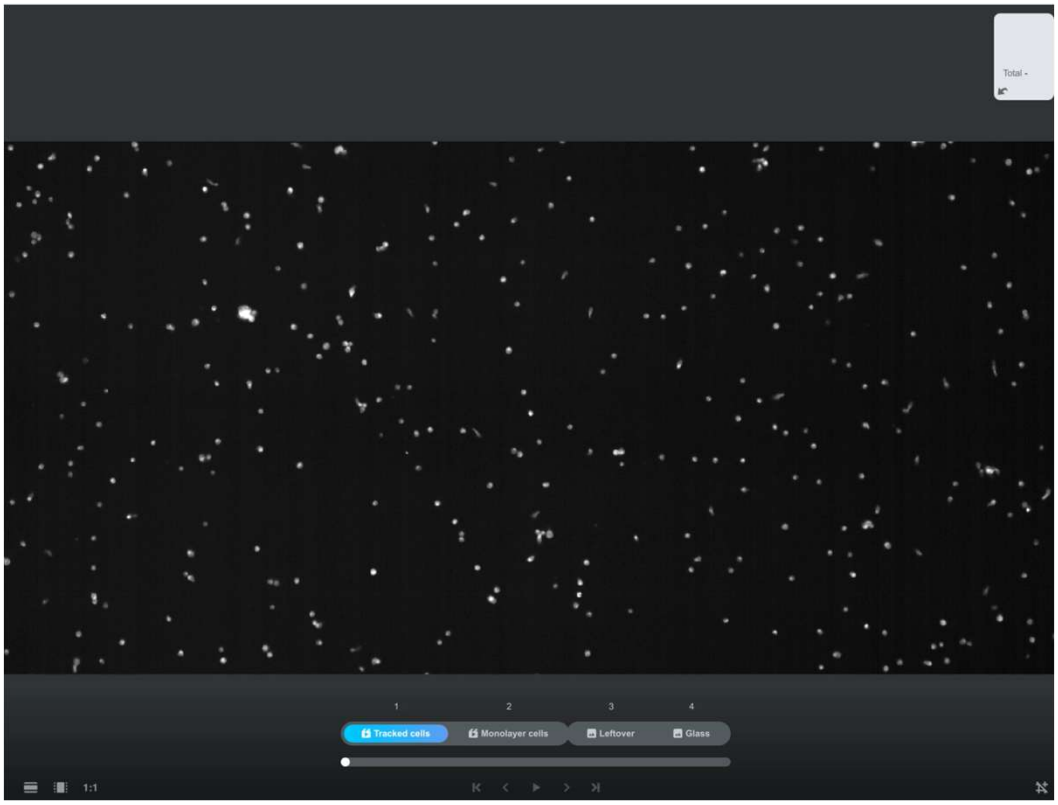
Applying acoustic forces to measure cell interaction



References
Sitters et al., Nature Methods 2015 – Acoustic technology principle | Kamsma et al., Cell Reports 2018 – CD4+ T cell adhesion to fibronectin

Measuring cell avidity with acoustic force and live-cell imaging

UTD Control

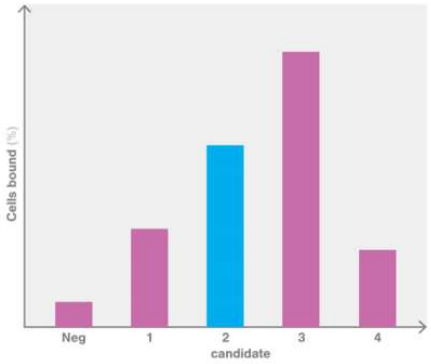


CD19 CAR-T

Four Key Applications of Cell Avidity

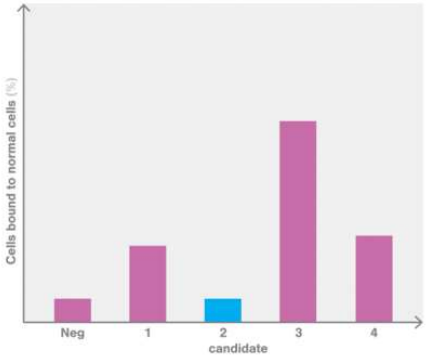
1. Potency

Measure antigen specific binding in the whole cellular context, which represents the initiating event in immune synapse formation.



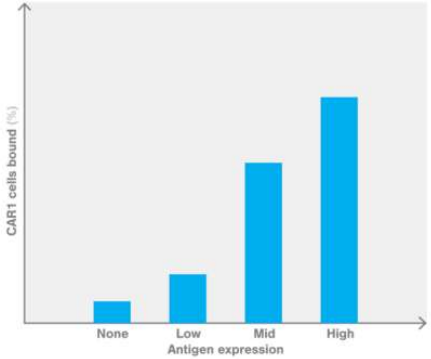
2. Specificity

Establish a quick and easy safety assay to determine the likelihood of off-target cellular binding.



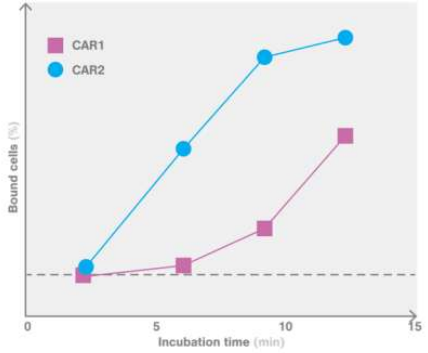
3. Sensitivity

Measure binding sensitivity across a variety of antigen concentrations to confirm the likelihood of downstream activation.



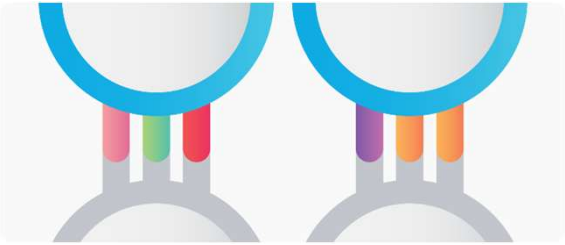
4. Kinetics

Measure binding kinetics on the cellular level to reveal deeper mechanistic insight.



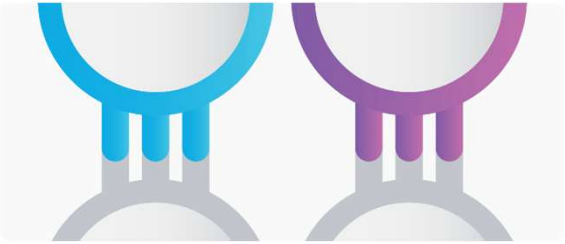
Improved Insights with Cell Avidity

Binder screening and optimization



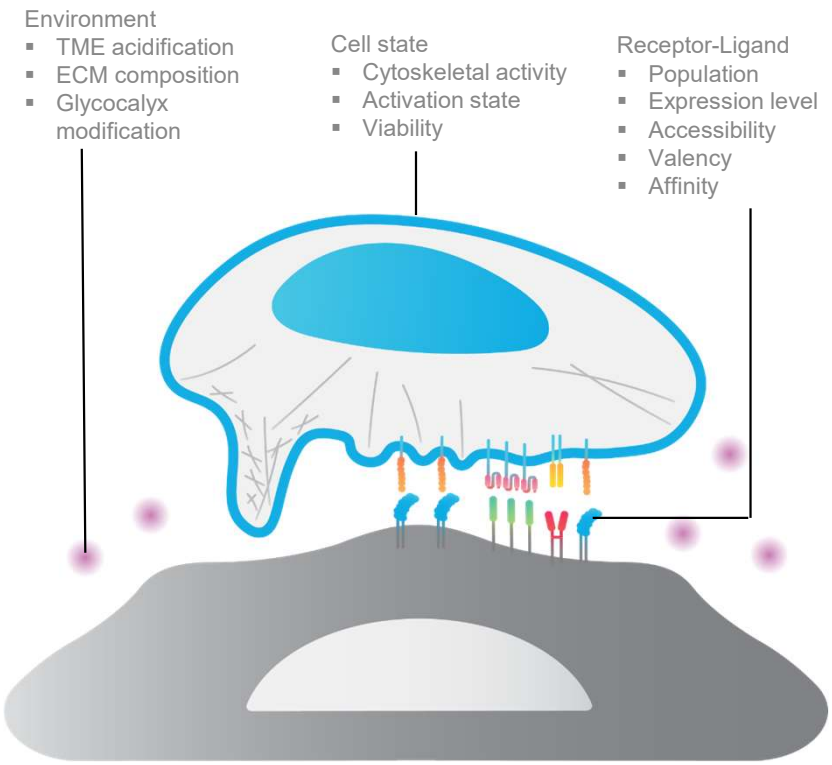
Ecto-domain modifications
Multi valent targeting

Donor selection and quality control

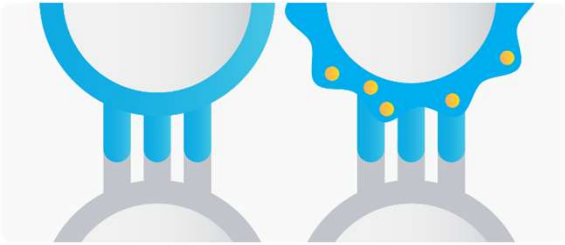


Sample specific binder proteomics
and cell activity

Cell-cell binding is governed by a complex and dynamic interplay between the multitude of receptor-ligand binding and cell activity in the context of the physiological environment

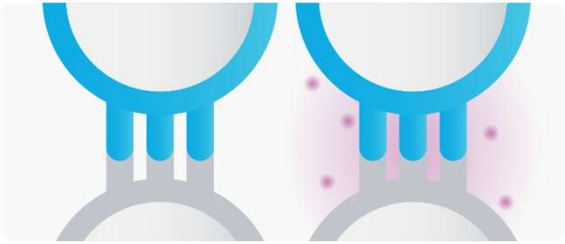


Effector functional modification



Immune checkpoint inhibition
Armouring / Chemokine receptors

Tumor microenvironment response



TME hypoxia and acidification
ECM binding and glycocalyx sensitivity

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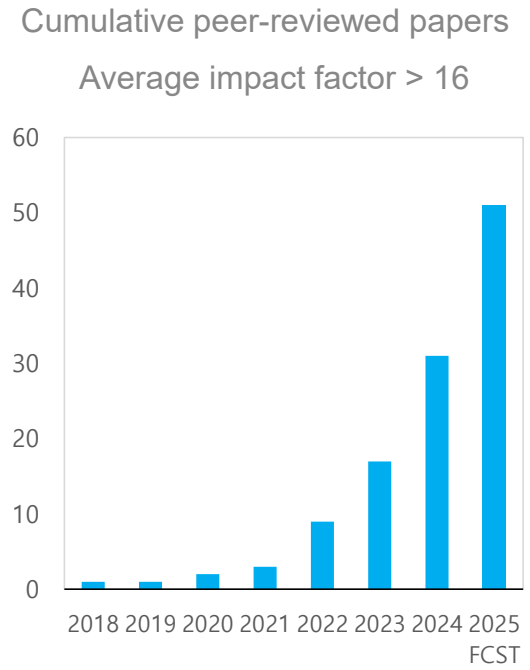
Cell Avidity enables rational design, leading to superior cell therapies

Group	Challenge	**Outcome**: *Cell Avidity Analysis **preeminently**...*
 Dr. Marco Ruella UPenn	Overcoming TME immunosuppression	...identified the BTLA-HVEM axis as a key T cell immunosuppressive mechanism. *(Guruprasad et al., Nat. Immunol., 2024)*
 Dr. Marcella Maus MGH	Overcoming immune evasion in TME	...showed the dual targeting mechanism of CAR-TEAM cells inducing binding to pancreatic cancer cells and cancer-associated fibroblasts. *(Wehrli et al., Clin. Cancer Res., 2024)*
 Dr. Wayne Marasco Dana Farber	Mitigating OTOT toxicity	… demonstrated selective high binding of a low affinity CAR to tumor cells over healthy cells, enabling the identification of a superior and safe CAR. *(Wang et al., Mol. Cancer, 2024)*
 Dr. David Weiner Wistar Institute	Improving half-life for a Cell Engager	...supported a rational design approach increasing plasma half-life of a cell engager by 30-fold while preserving high avidity binding. *(O'Connell et al., J. Immunother. Cancer, 2024)*
 Dr. Stephan Gottschalk St. Jude's Children	Ensuring potency for CAR-NK therapy	...revealed the mechanism of CAR-NK cells with an intracellular PDZ modification, aiding informed decisions in translational development. *(Chockley et al., Nat. Biotech, 2023)*

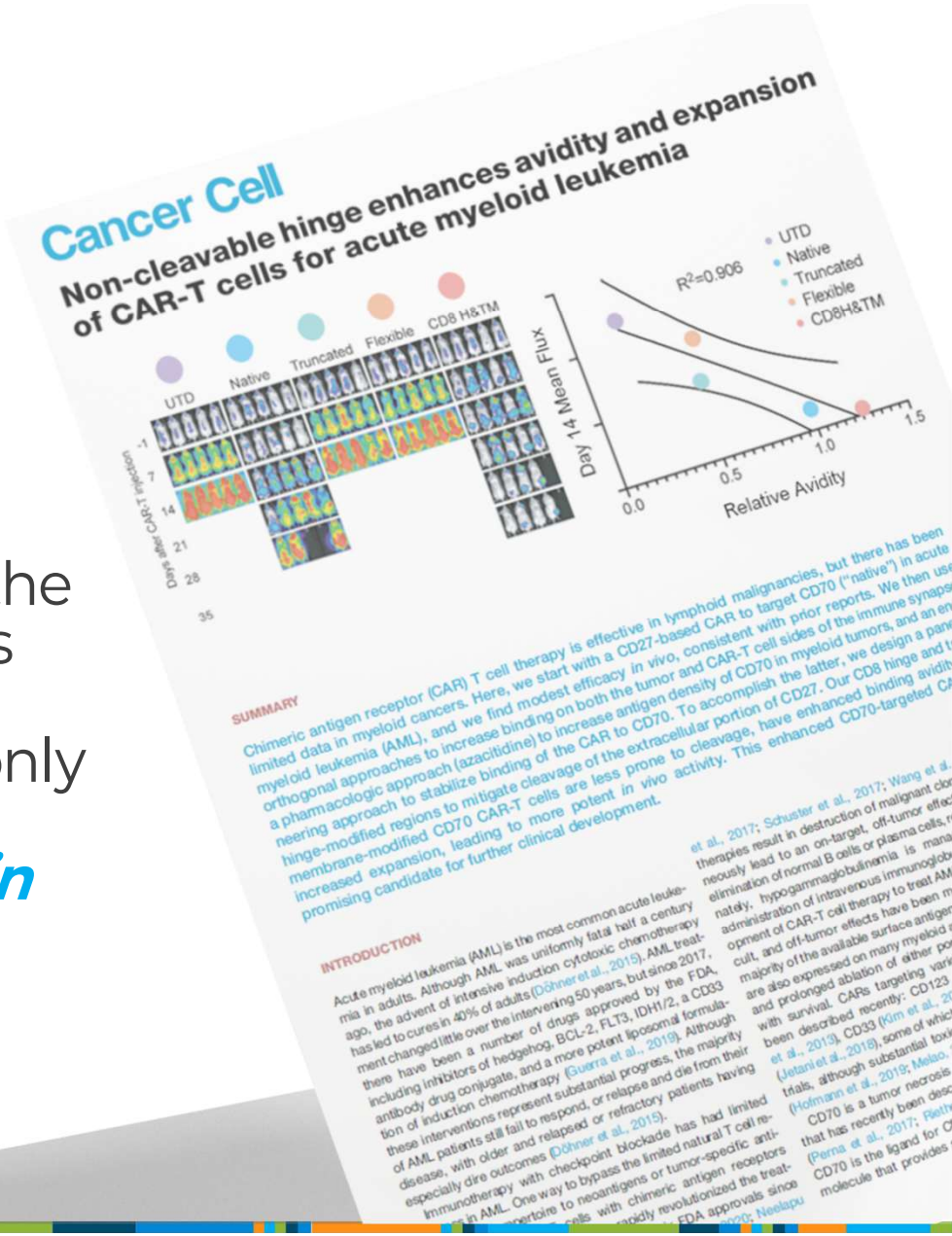


Director of Cellular Immunotherapy

Harvard Medical School

Massachusetts General Hospital

"Interestingly, of the pre-clinical assays used to compare CAR constructs, only binding **avidity correlated with *in vivo* results**."


Cancer Cell

Non-cleavable hinge enhances avidity and expansion of CAR-T cells for acute myeloid leukemia

UTD Native Truncated Flexible CD8 H&TM

Days after CAR-T infection

Day 14 Mean Flux

R^2=0.906

Relative Avidity

SUMMARY

Chimeric antigen receptor (CAR) T cell therapy is effective in lymphoid malignancies, but there has been limited data in myeloid cancers. Here, we start with a CD27-based CAR to target CD70 ("native") in acute myeloid leukemia (AML), and we find modest efficacy in vivo, consistent with prior reports. We then use orthogonal approaches to increase binding on both the tumor and CAR-T cell sides of the immune synapse: a pharmacologic approach (azacitidine) to increase antigen density of CD70 in myeloid tumors, and an engineering approach to stabilize binding of the CAR to CD70. To accomplish the latter, we design a panel of hinge-modified regions to mitigate cleavage of the extracellular portion of CD27. Our CD8 hinge and transmembrane-modified CD70 CAR-T cells are less prone to cleavage, have enhanced binding avidity, increased expansion, leading to more potent in vivo activity. This enhanced CD70-targeted CAR is a promising candidate for further clinical development.

INTRODUCTION

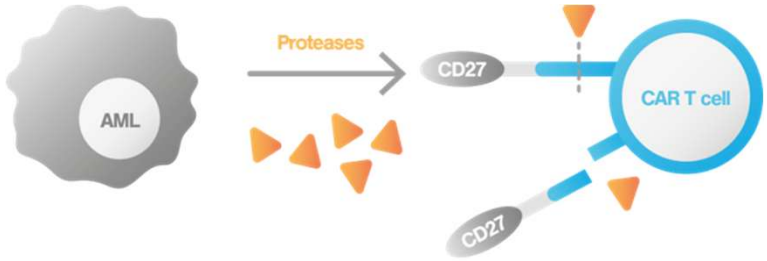
Acute myeloid leukemia (AML) is the most common acute leukemia in adults. Although AML was uniformly fatal half a century ago, the advent of intensive induction cytotoxic chemotherapy has led to cures in 40% of adults (Döhner et al., 2015). AML treatment changed little over the intervening 50 years, but since 2017, there have been a number of drugs approved by the FDA, including inhibitors of hedgehog, BCL-2, FLT3, IDH1/2, a CD33 antibody drug conjugate, and a more potent liposomal formulation of induction chemotherapy (Guerra et al., 2019). Although these interventions represent substantial progress, the majority of AML patients still fail to respond, or relapse and die from their disease, with older and relapsed or refractory patients having especially dire outcomes (Döhner et al., 2015).

Immunotherapy with checkpoint blockade has had limited success in AML. One way to bypass the limited natural T cell repertoire to neoantigens or tumor-specific antigens is with chimeric antigen receptors (CARs), which have rapidly revolutionized the treatment of [illegible] cells with chimeric antigen receptors ... FDA approvals since ... (Neelapu et al., 2017; Schuster et al., 2017; Wang et al., ... therapies result in destruction of malignant cells ... neously lead to an on-target, off-tumor effect ... elimination of normal B cells or plasma cells, ... nately, hypogammaglobulinemia is managed ... administration of intravenous immunoglobulin ... opment of CAR-T cell therapy to treat AML ... cult, and off-tumor effects have been mitigated ... majority of the available surface antigens ... are also expressed on many myeloid cells ... and prolonged ablation of either population ... with survival. CARs targeting various ... been described recently: CD123 (Kim et al., 2018; ... et al., 2013), CD33 (Kim et al., 2018), some of which ... (Jetani et al., 2018; Mehta ... trials, although substantial toxicity ... (Hofmann et al., 2019; Mehta ... CD70 is a tumor necrosis ... that has recently been described ... (Perna et al., 2017; Riether ... CD70 is the ligand for CD27 ... molecule that provides ...

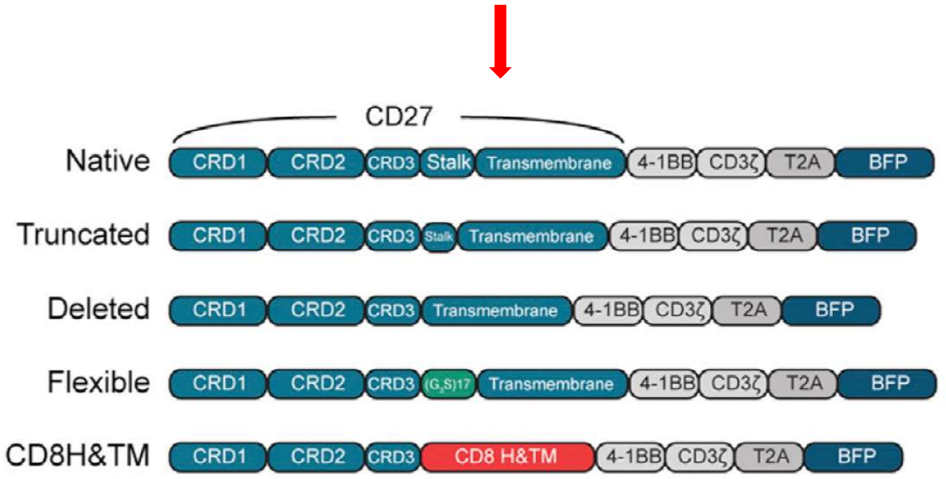
References
Leick et al., Cancer Cell 2022

Avidity Enhancement Predicts AML Eradication

Problem: Targeting CD70 in AML results in cleavage of CD27 scFv and limited in-vivo response



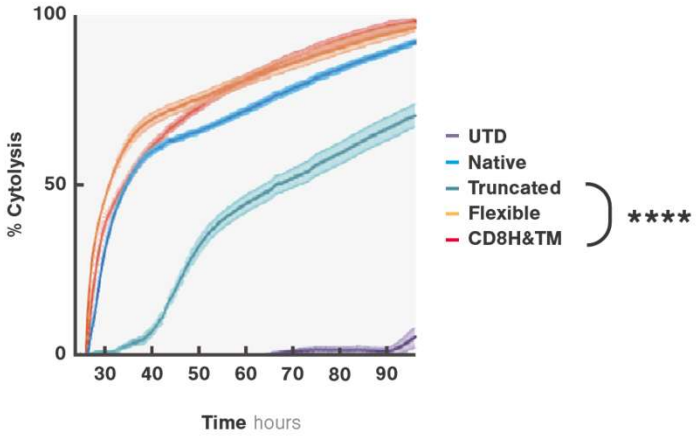
Solution: design CARs without cleavage site, increasing cell avidity



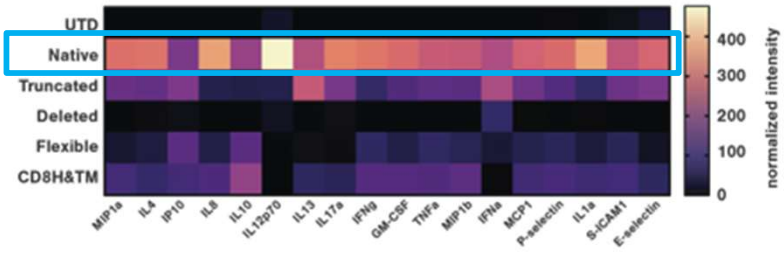
References
Leick et al., Cancer Cell 2022

Killing & Cytokine Readouts Are Insufficient for Candidate Selection

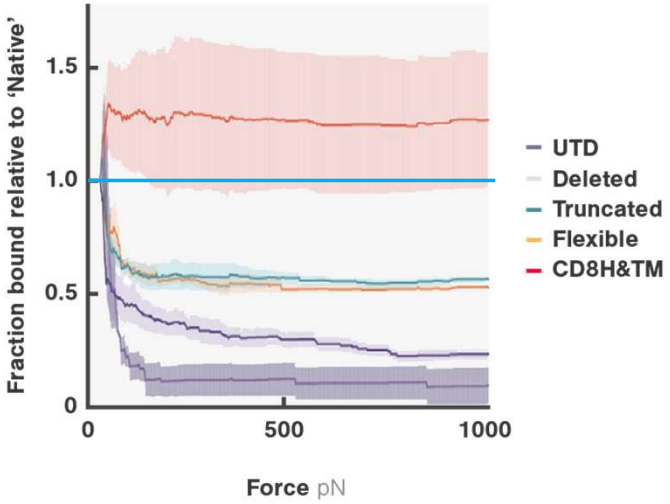
Serial restimulation assay did not distinguish between CAR variants



cytokine secretion selected the Native CAR as best candidate

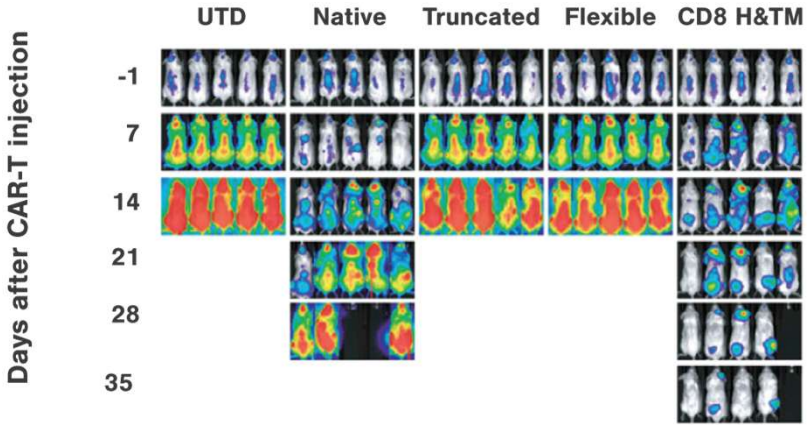


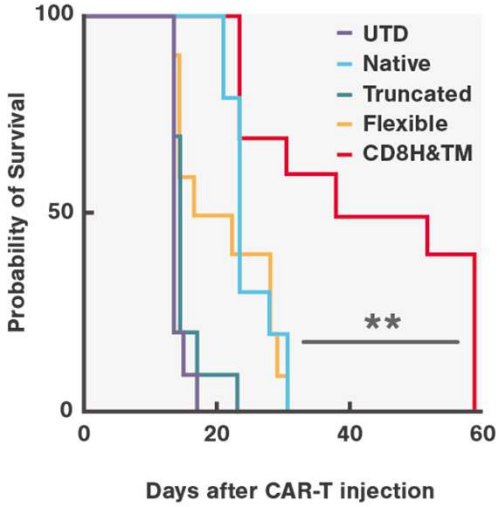
Avidity data shows the CD8H&TM CAR outperforming all others



References
Leick et al., Cancer Cell 2022

Avidity-Enhanced CAR Doubles Survival





References
Leick et al., Cancer Cell 2022

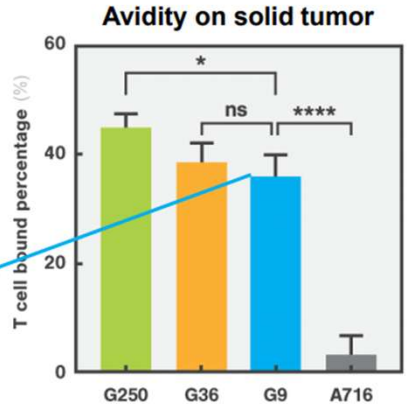
Avidity Analysis Confirms Safe CAR-T Binding Post Affinity Tuning

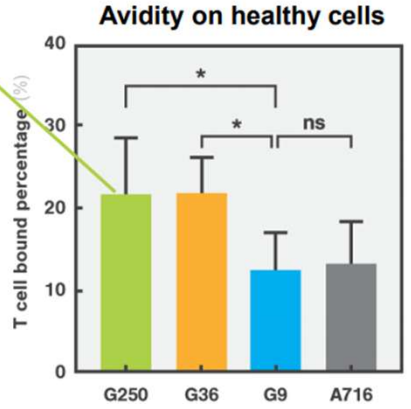
Dr. Yufei Wang
Sr. Postdoctoral researcher
Dana Farber Cancer Institute

Hypothesis

Incorporating a lower-affinity scFv domain will allow sufficiently potent binding of tumor cells, while mitigating on-target off-tumor bindings.

Avidity analysis demonstrates safe CAR-T binding to healthy cells





Outcome

G9 CAR-T product shows strong and potent binding to solid tumor cells, comparable to G250. In addition, as opposed to the clinically toxic G250, G9 anti-CAIX CAR-T demonstrates low and safe binding to healthy cells

Rhino Bio, Inc.

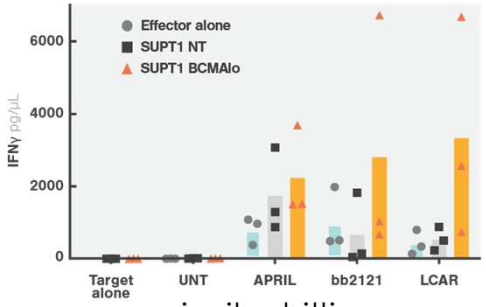
Wang et al., Mol. Cancer, 2024

LUMICKS

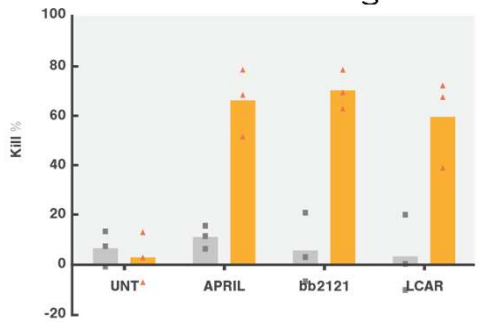
Current in vitro assays do not always predict clinical success

Sound *in vitro* results

similar activation

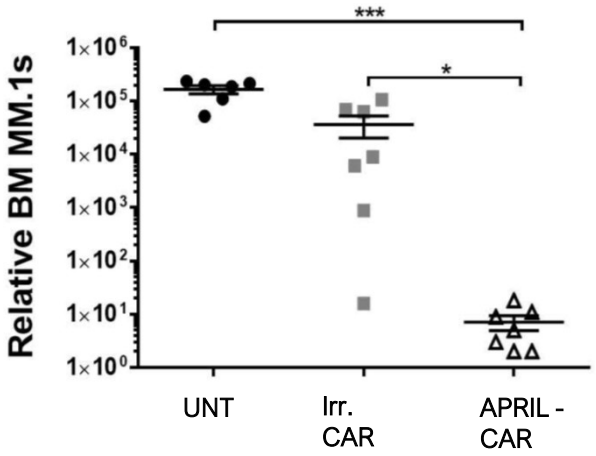


similar killing

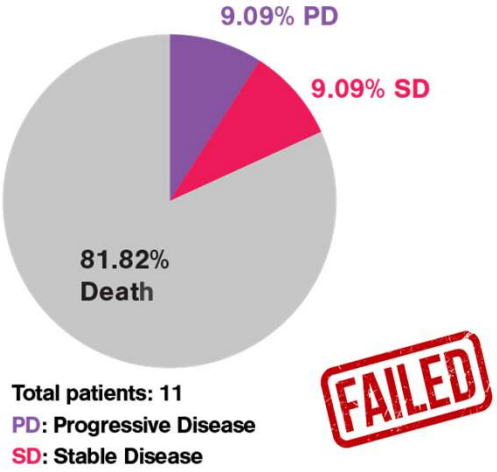


Passes *in vivo* tests

effective killing

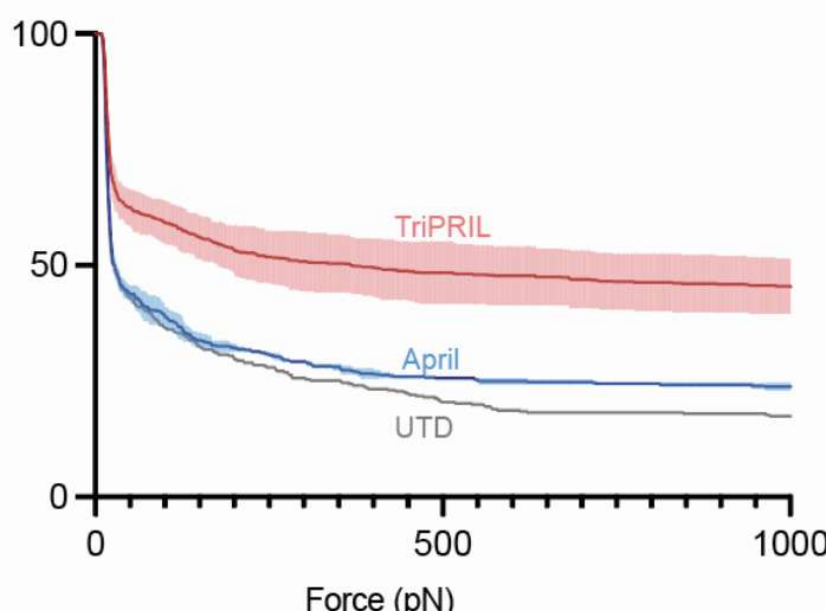


Fails in clinical trials



Lee et al, JITC, 2023

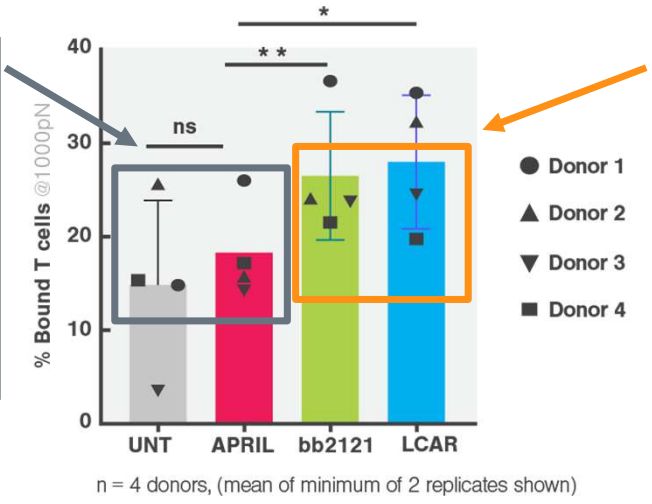
Preventing Trial Failure with Cell Avidity



Leick *et al*., Cancer Cell 2022

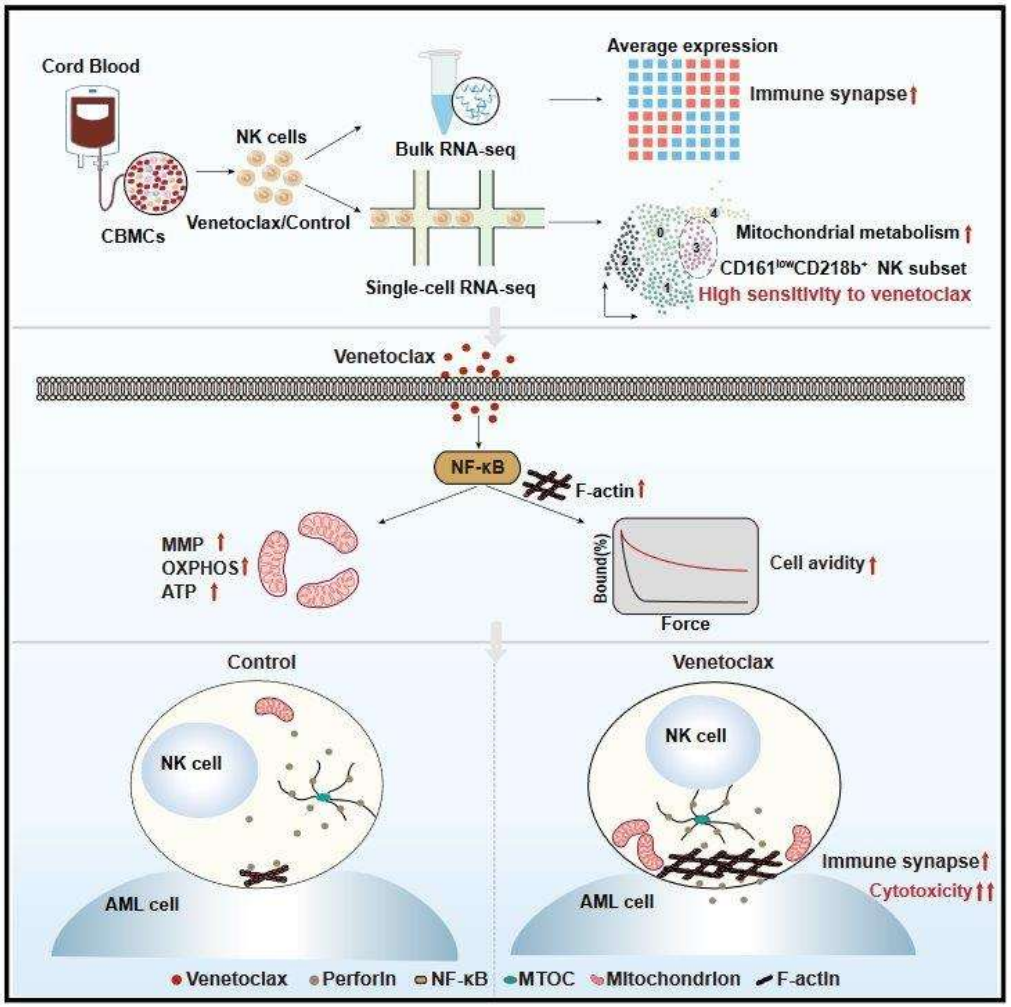
Despite good in vitro and in vivo functional data, APRIL CAR showed low avidity compared to other FDA approved CAR T cells

APRIL CAR displayed low avidity. No significant difference from negative control



FDA approved CAR-T cell from Bristol Myers Squibb & Janssen showed high avidity and good response in the clinic

Venetoclax enhances NK cell efficacy through increased Cell Avidity



Problem

Venetoclax, an FDA-approved drug, can boost NK cell function and killing in vivo; however, the mechanism behind this phenomenon is currently not understood.

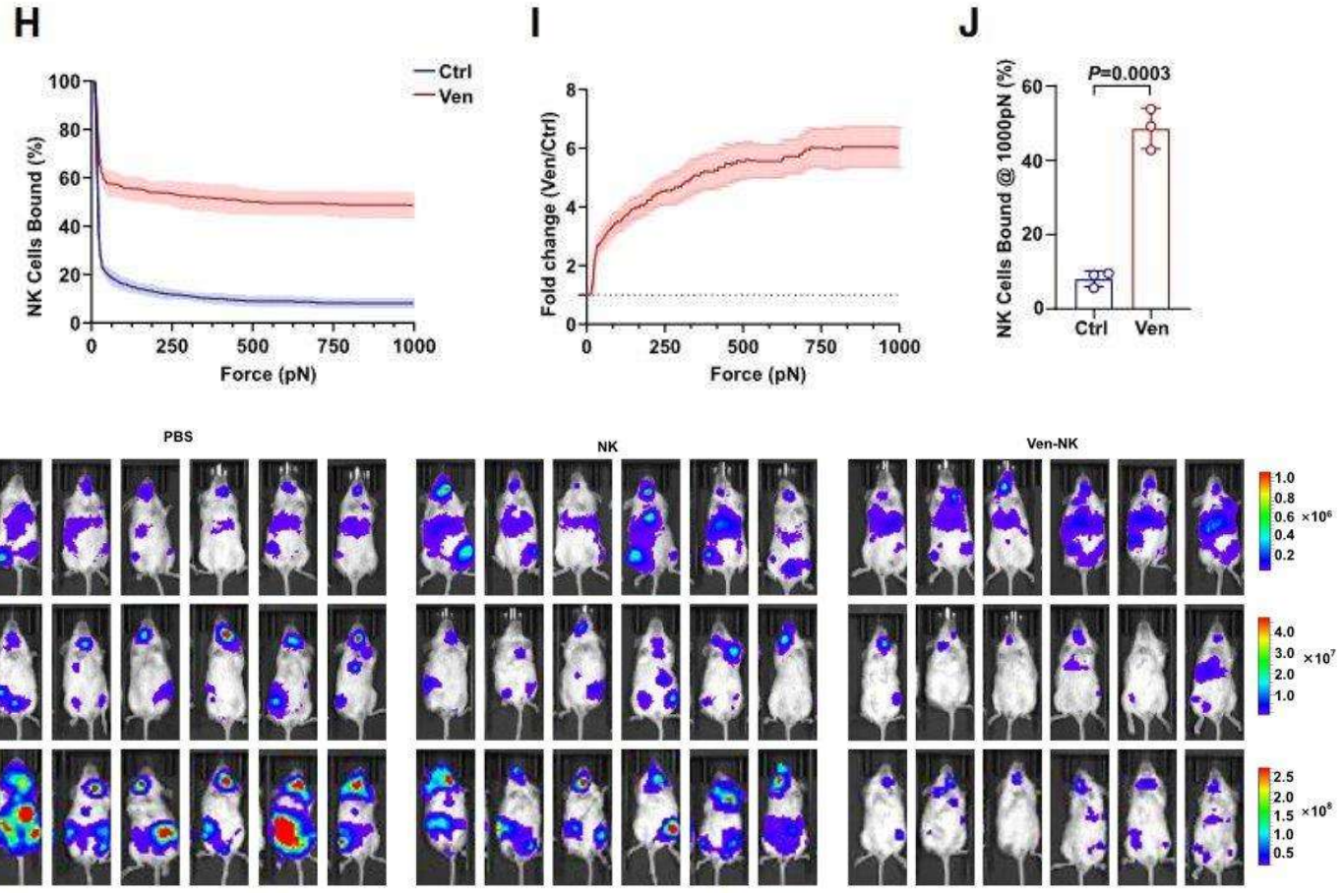
Hypothesis

Venetoclax treatment enhances NK cell-mediated killing of AML cells by increasing cellular avidity due to immunomodulatory effects.

Dr. Fang Ni
Principal Investigator
Institute of Immunology
USTC China

 Wang et al., Cell Rep. Med., 2024

Increased NK Cell Avidity Improves Tumor Control In Vivo



Outcome

Venetoclax increased cell avidity, enhancing NK cell target cell interaction, which boosted NK cell function, opening avenues for its therapeutic potential in various cancers.

Dr. Fang Ni
Principal Investigator
Institute of Immunology
USTC China

Cell Avidity: a Swiss army knife for next generation binding

Cell Avidity

- Immune-oncology
- Autoimmunity
- Inflammatory disease
- Infectious disease

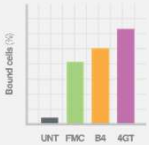
Cell-cell binding

Cell Therapy

Cell-surface binding

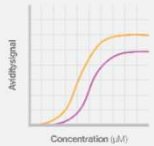
Biologics
Gene therapy

Binding potency



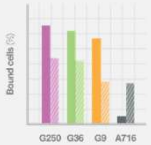
Avidity ranking & Cell characterization

Binding sensitivity



Titration assay for CE / Peptide / AB

Binding specificity

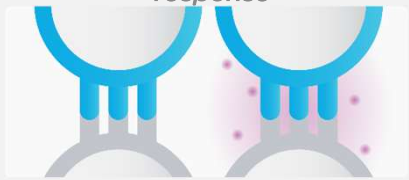


Monolayer safety screening

Binder screening and optimization

Effector functional modification

Tumor microenvironment response



Donor selection and quality control

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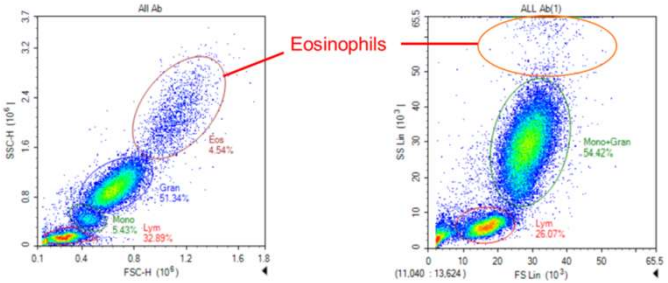
LUMICKS

Agilent Novocyte Flow Cytometer

Spectral

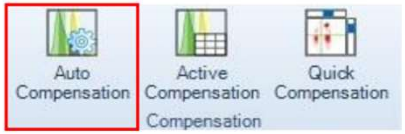
Conventional

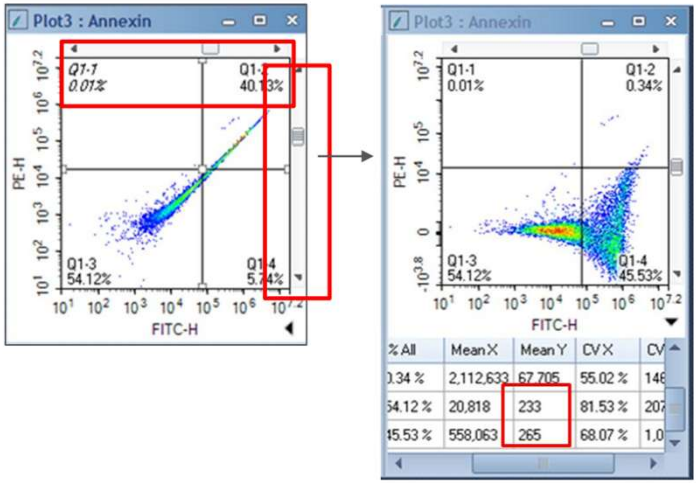
10^7 dynamic range



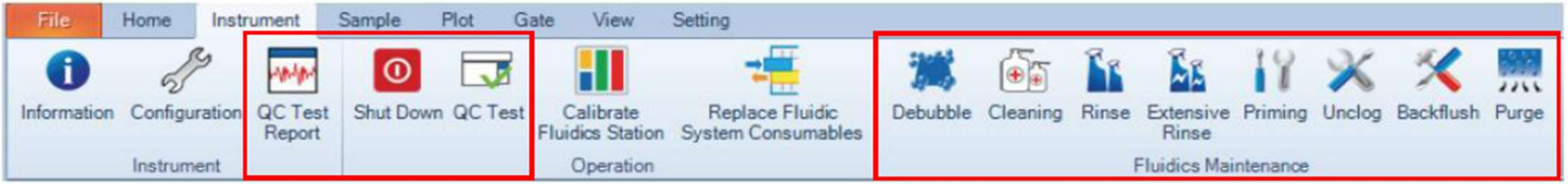
NovoCyte Competitor

Auto compensation





QC test & Fluidics Maintenance



Thank You

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