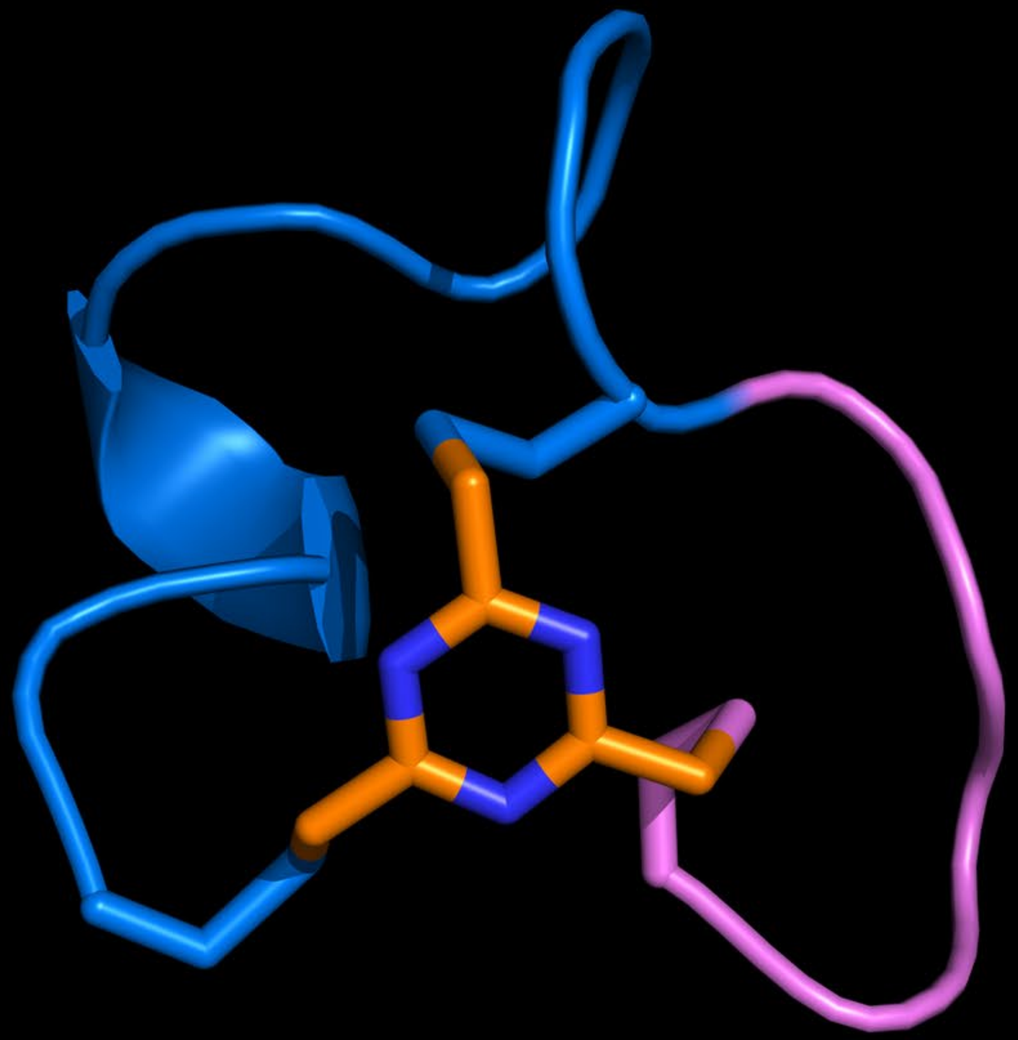
Bicycle Therapeutics: Precision-guided immune agonism for the treatment of cancer

Phil Brandish, VP Immuno-oncology

Immuno UK meeting, London
September 30th, 2022



Bicycle®

Presentation outline

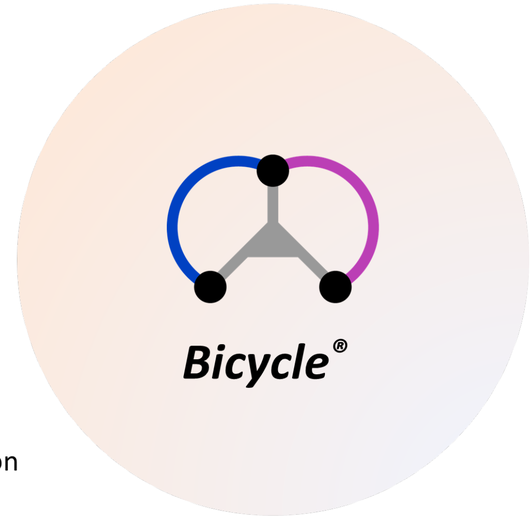
- Introduce the *Bicycle®* platform and its application to cancer therapy
- Discuss the discovery, development and MOA of precision-guided CD137 agonists including BT7480
- Discuss NK cell engagers and application of the *Bicycle®* technology in this area

Bicycles are short peptides chemically constrained with a central scaffold

Short linear peptide

+

Scaffold
Chemical modification with scaffold

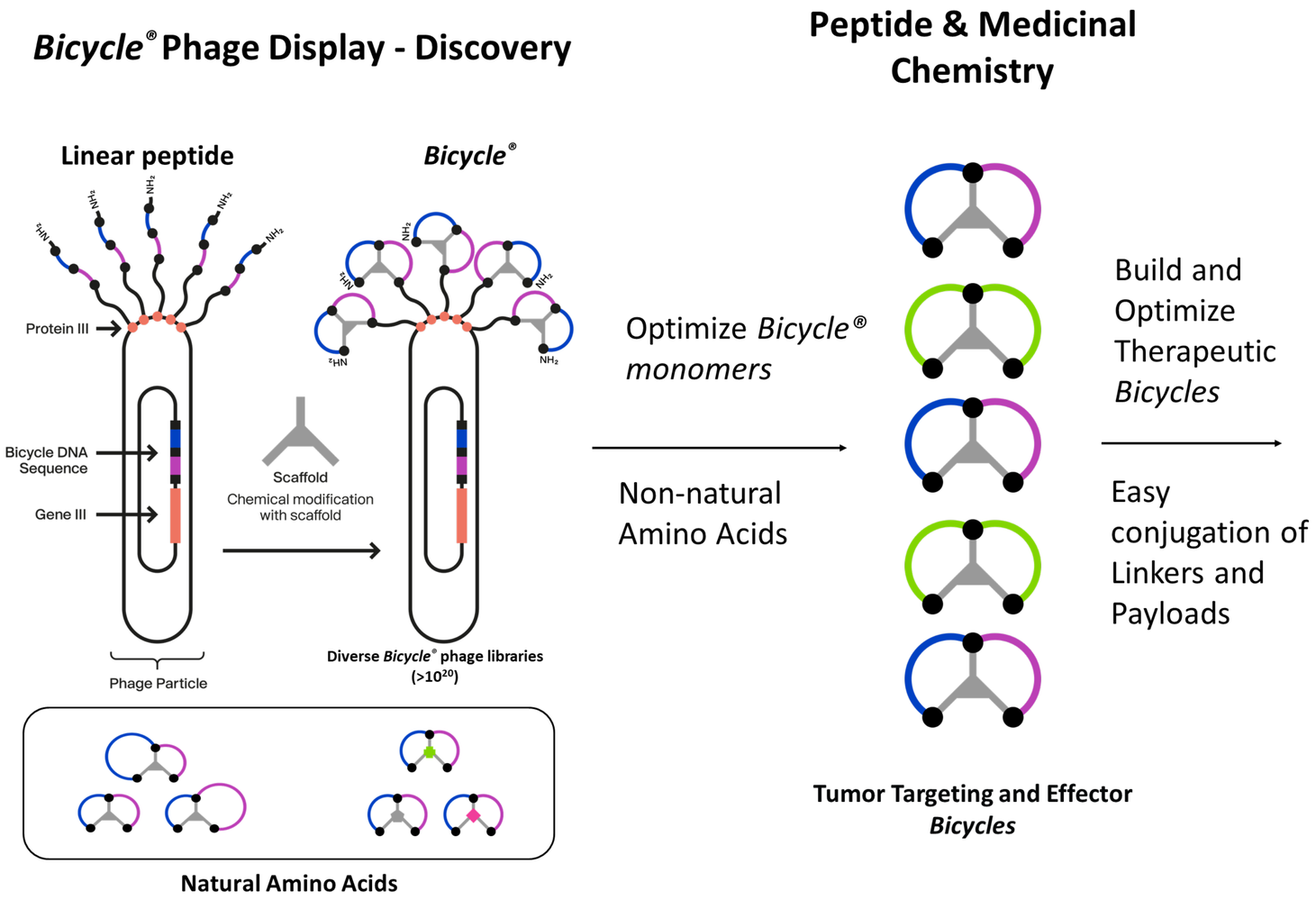


Based on the work of Greg Winter to define the minimal mAb pharmacophore

- 2018 Nobel Prize in Chemistry
- Founded Bicycle Therapeutics

	Bicycle®
Small size	**Yes 1.5 to 2 kDa**
Specificity	**High**
Chemical synthesis (NCEs)	**Yes**
Rapid tissue penetration	**Yes**
Complex protein targets druggable	**Yes**
Route of elimination	**Renal**

How *Bicycles* are discovered and why they work



Cyclization on the surface of the phage means we screen for the constrained 3D structure, not the sequence

Having been discovered while attached to a phage particle, conjugation to a payload, including other *Bicycles*, rarely impairs binding to target

Switching to chemical synthesis after screening introduces non-natural amino acids & leverages enormous proprietary data sets

First application in cancer – delivery of cytotoxic agents maximizing tumor concentration while minimizing exposure in the periphery

Property	BTC	ADC	Importance
Tumor penetration	✓	?	Access to site of action
Tumor retention	✓	?	Maintenance at site of action, lower total body burden
Short systemic exposure	✓	✗	Minimizes toxicity, enhances combinability
Reduced liver metabolism	✓	✗	Improved safety profile
Renal elimination	✓	✗	Improved safety profile
Flexible dosing	✓	✗	Tailored dosing regimen minimizing toxicity

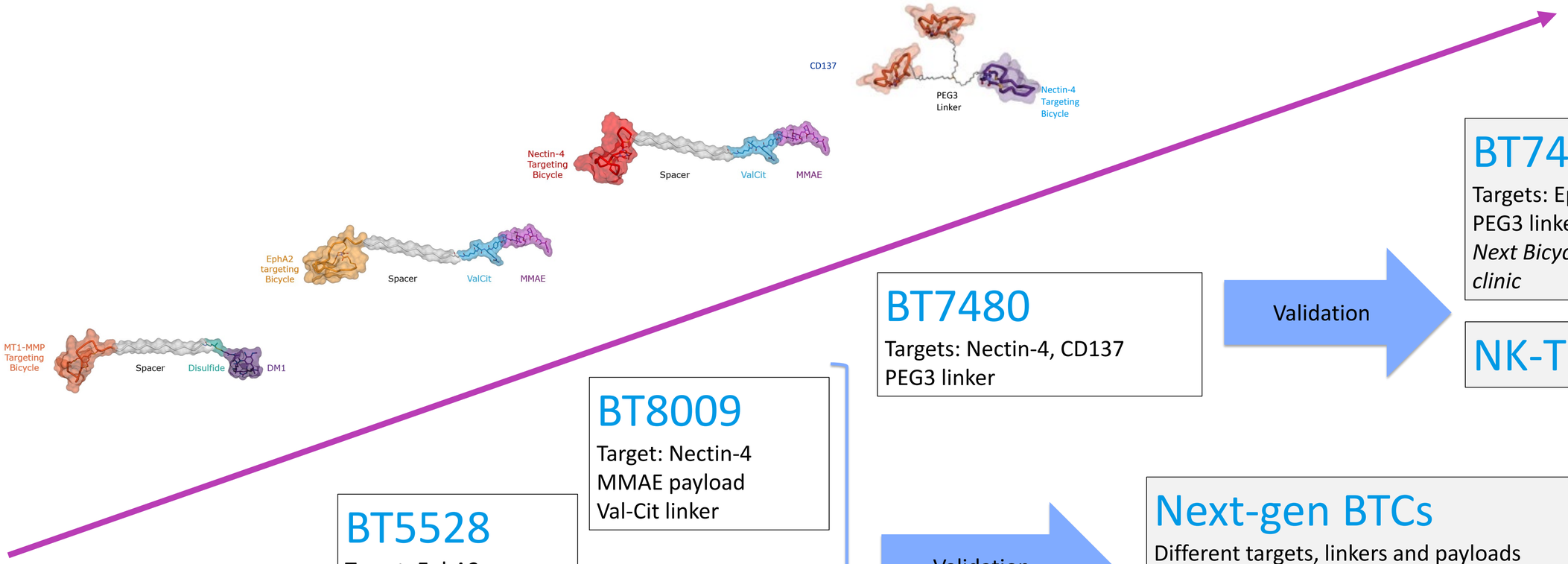
The properties that make *Bicycles* great for toxin delivery also make them great for immune agonism, but for different reasons

- In the body, activating signals (agonists) are local, rapid, and then stop
  - Neurotransmitters
  - Cytokines
  - Stress hormones

- Sustained (pathologic) signaling leads to desensitization and dysregulation

Bicycles are precision-guided (local), distribute in the tissues quickly (rapid), and are cleared rapidly from the body (stop)

Elevating the platform



BT1718
Target: MT1-MMP
DM1 payload
Disulfide linker

BT5528
Target: EphA2
MMAE payload
Val-Cit linker

BT8009
Target: Nectin-4
MMAE payload
Val-Cit linker

BT7480
Targets: Nectin-4, CD137
PEG3 linker

Validation

BT7455
Targets: EphA2, CD137
PEG3 linker
Next Bicycle TICA™ scheduled for clinic

NK-TICA™

Validation

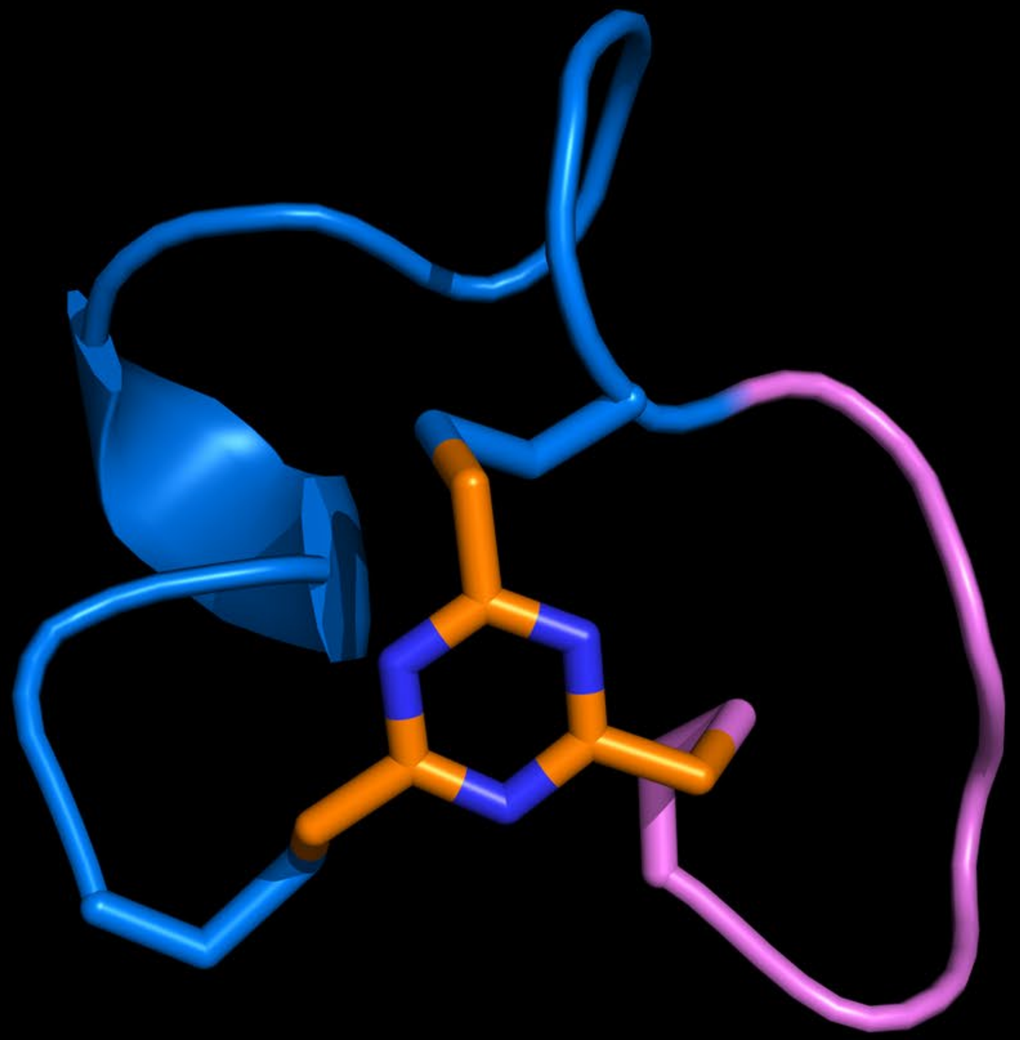
Next-gen BTCs
Different targets, linkers and payloads

Radiopharm

FIH 2018 2019 2020 2021 2023 and beyond

Bicycle® precision-guided immune activation

Immune cell receptor = CD137



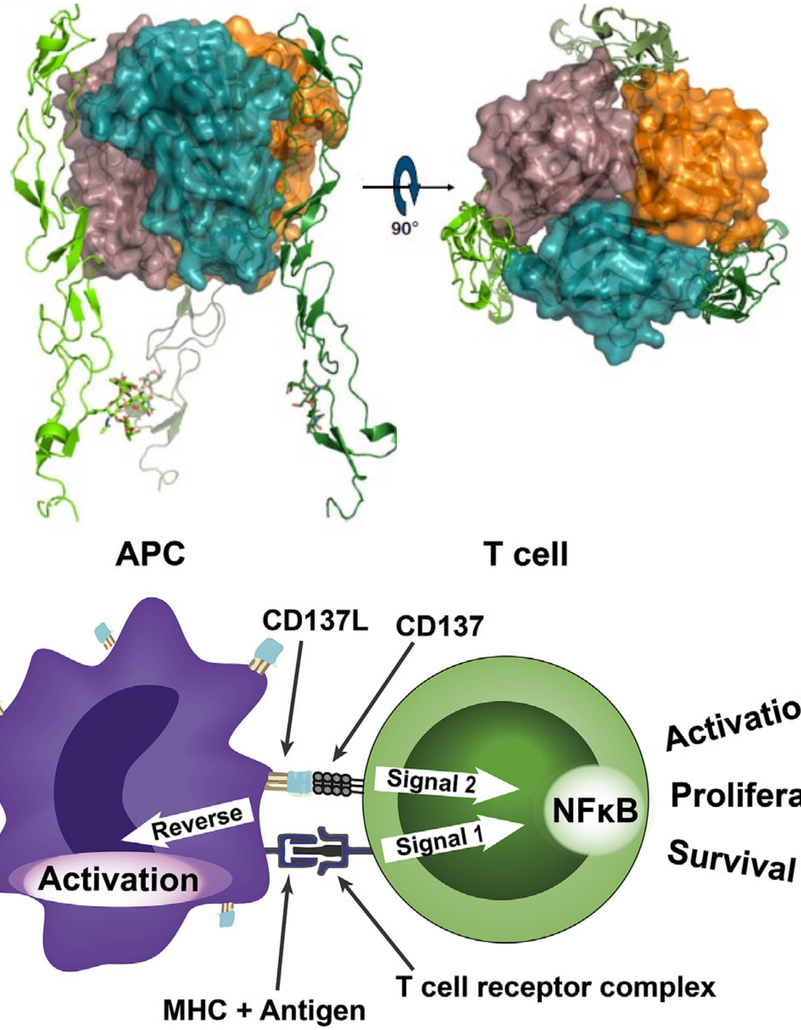
Bicycle®

CD137 (4-1BB) is an immune co-stimulatory receptor with high therapeutic potential in cancer

- CD137 is expressed on activated immune cells – signaling enhances function and survival, prevents anergy
- CD137 ligand expressed by APCs provides a co-stimulatory signal to T cells and NK cells – potential in antitumor immunity
- Sustained activation leads to exhaustion and AICD – transient, localized action may be the optimal approach
- Urelumab – anti-CD137 agonist mAb – some clinical activity but liver toxicity precluded development

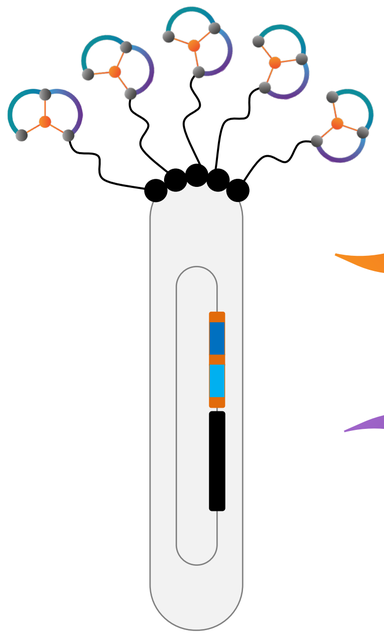
Many agents in development now – none meet design goals dictated by the biology – we sought to address this using the *Bicycle®* platform:

- Activity localized to the tumor – potentiate immune activation
- Rapid onset of action and controllable duration of action
- No Fc interactions to avoid liver toxicity



Yonezawa (2015); Melero (2008) TiPS 29, 383; Melero (2007) Nat. Immunol 3, 682; Wilcox (2004) Blood 103, 177; Wilcox (2002) J. Immunol. 169, 4230; Gomes-Silva (2017) Cell Rep. 21, 17; Segal (2016) Clin. Cancer Res. 23, 1929; Zheng – SITC2020 abstract 812; Chin (2018) Nat. Comm. 9, 4679; Soderstrom (2018) Atherosclerosis 272, 66

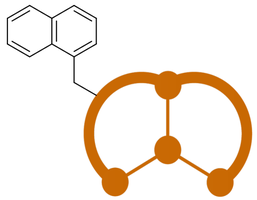
CD137 and Nectin-4 *Bicycles*: discovery and optimization by phage display and chemistry



CD137

K_D = 1400 nM

K_D = 67 nM



K_D = 5 nM

Phage Hits

-ID sequence families
-Define initial pharmacophores

Phage Optimized

-Focused libraries
-Optimize natural AAs
-Scaffold/Loop symmetry

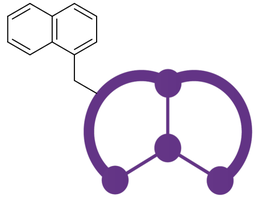
Chemistry Optimized

-Non natural amino acids
-Tune affinity and stability
-Balanced phys chem properties

Nectin-4

K_D = 508 nM

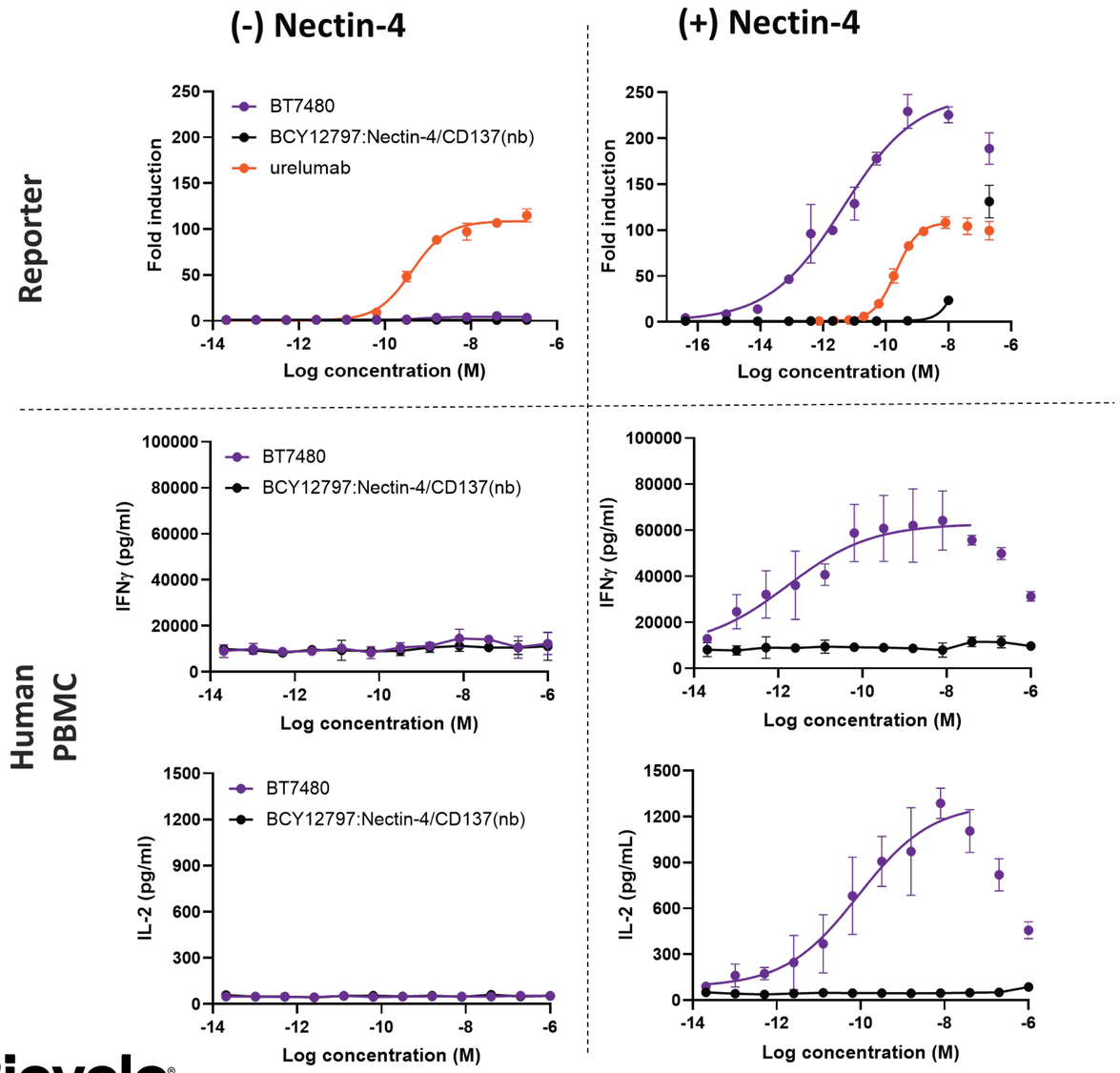
K_D = 80 nM

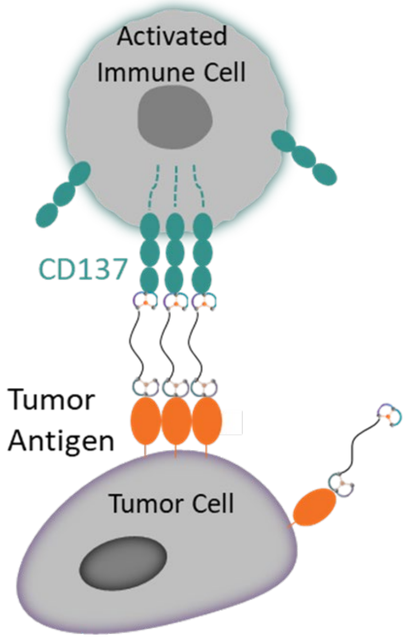


K_D = 1 nM

Bicycle®

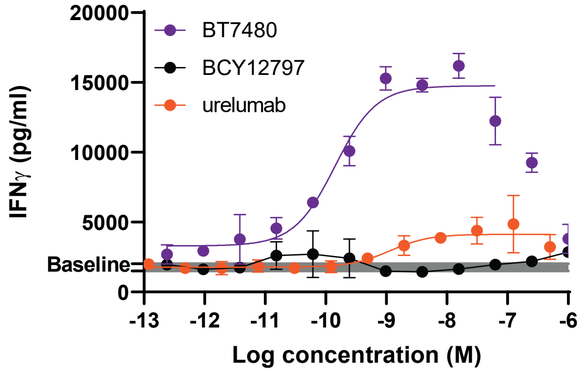
BT7480 functional activity is dependent on Nectin-4 in cell-based assays *in vitro*



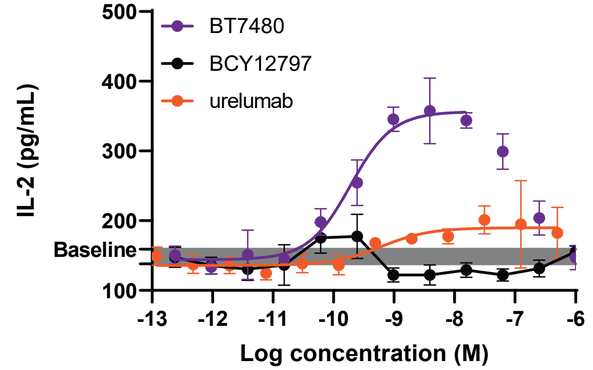


4T1 cells ± Nectin-4

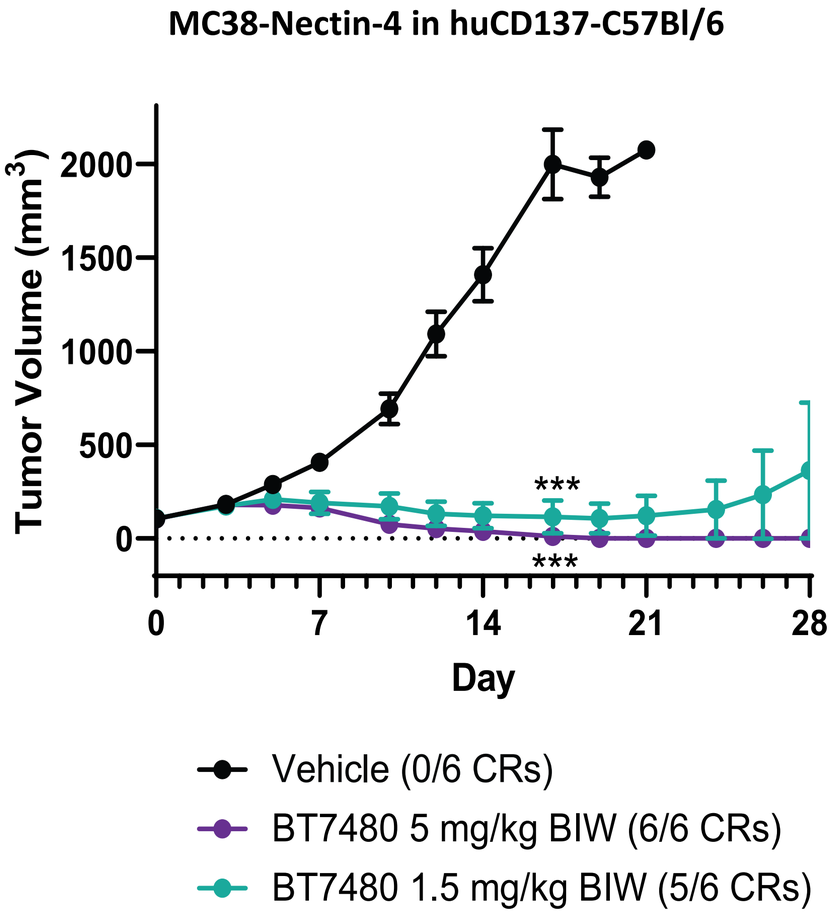
HT1376 bladder tumor cells



EC_{50}
0.37±0.23 nM (IL-2)
0.22±0.12 nM (IFNγ)

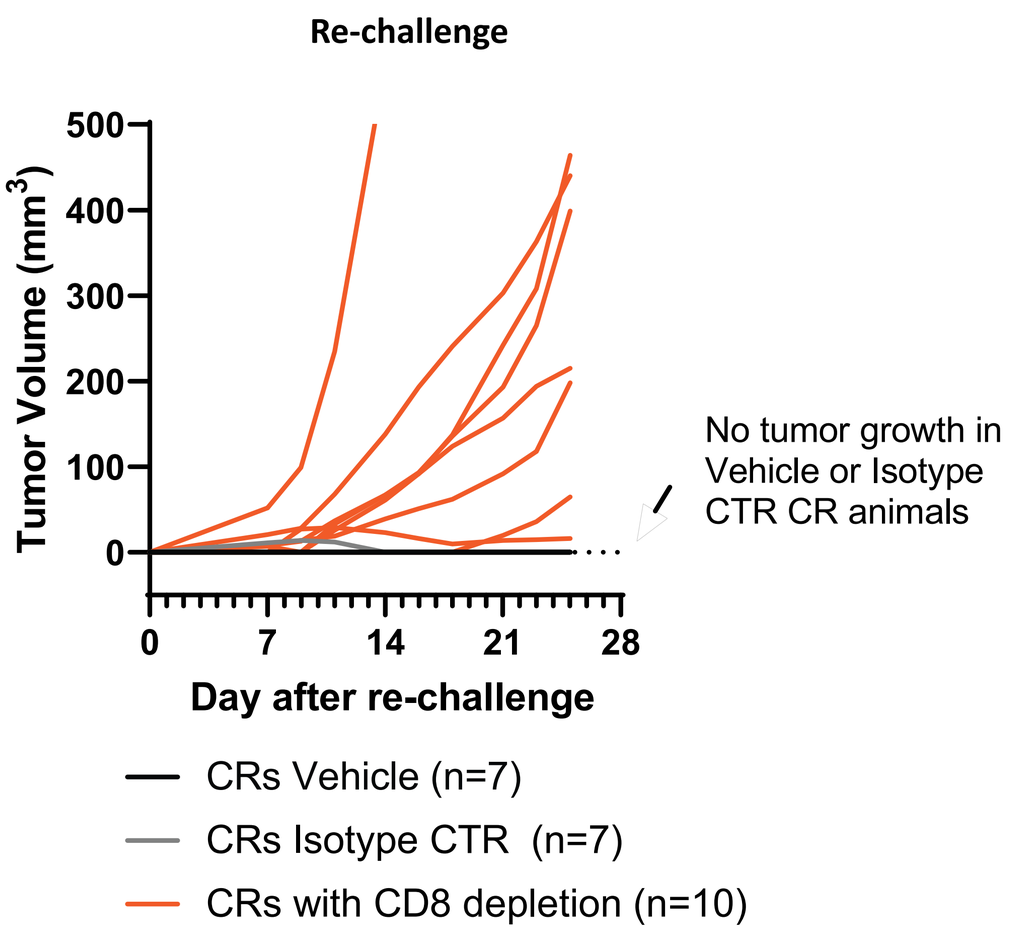


BT7480 induces complete responses and memory *in vivo* – mouse syngeneic MC38 tumor model

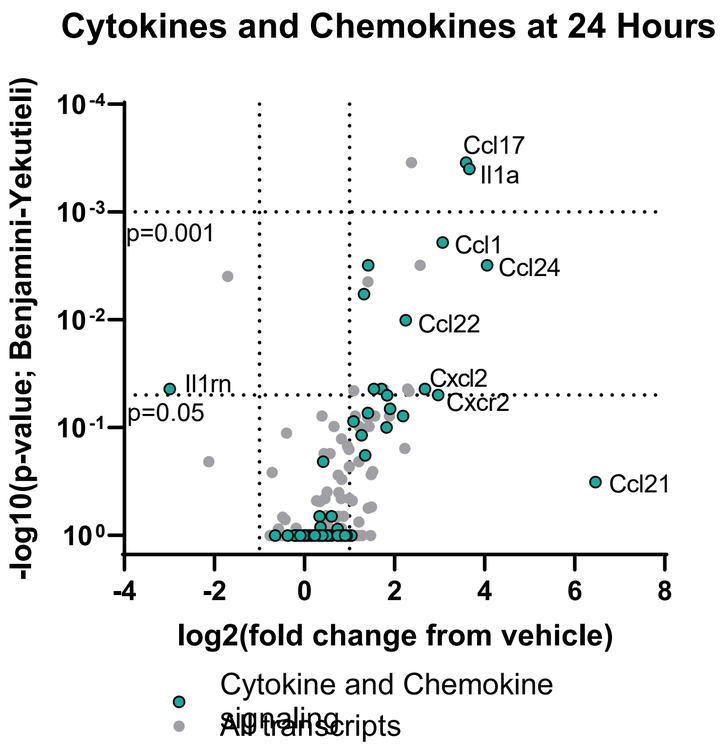


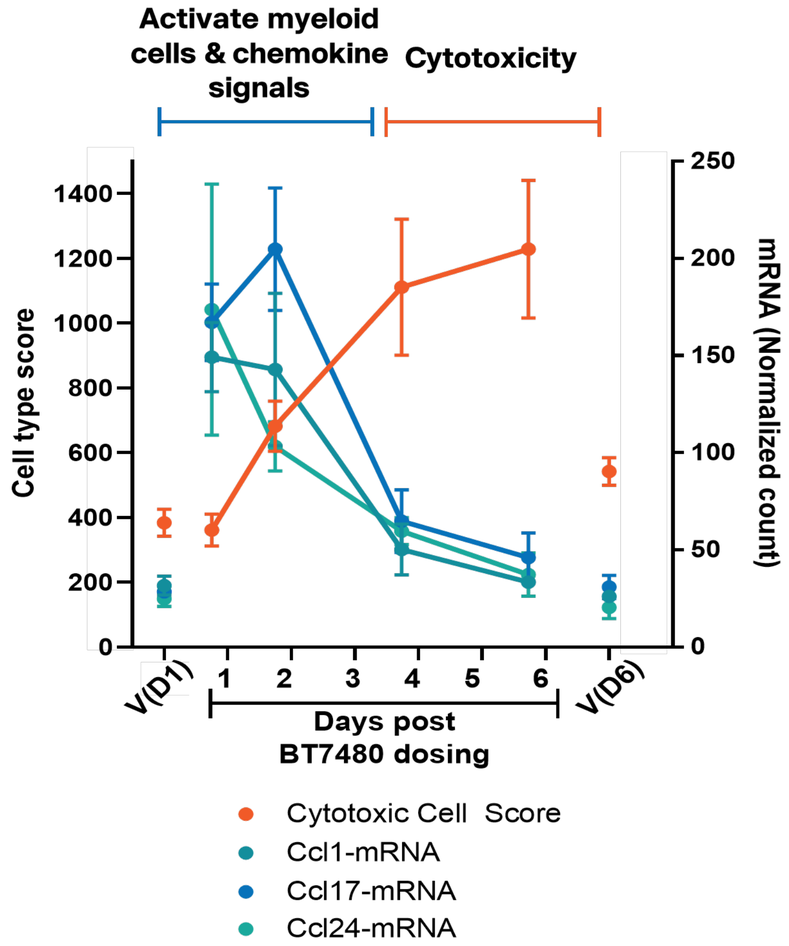
***p<0.001 Mixed effects analysis with Tukey's post test, days 0–17

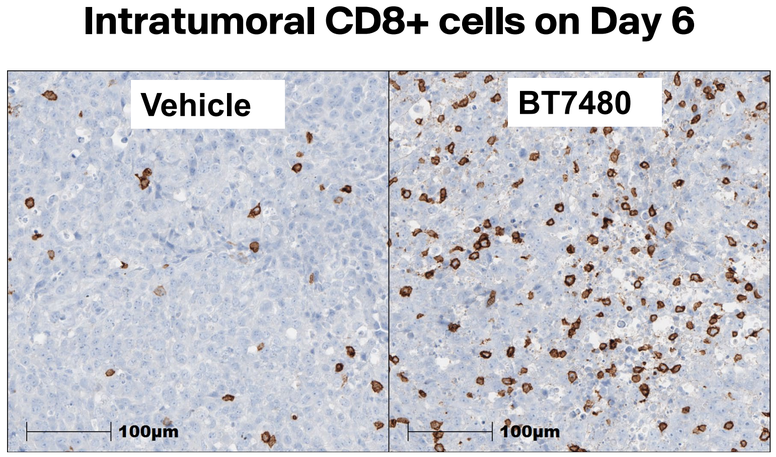
Day 59



Transcriptional analysis in mouse mc38 tumor model revealed an unanticipated, rapid burst of T cell chemotactic cytokine production



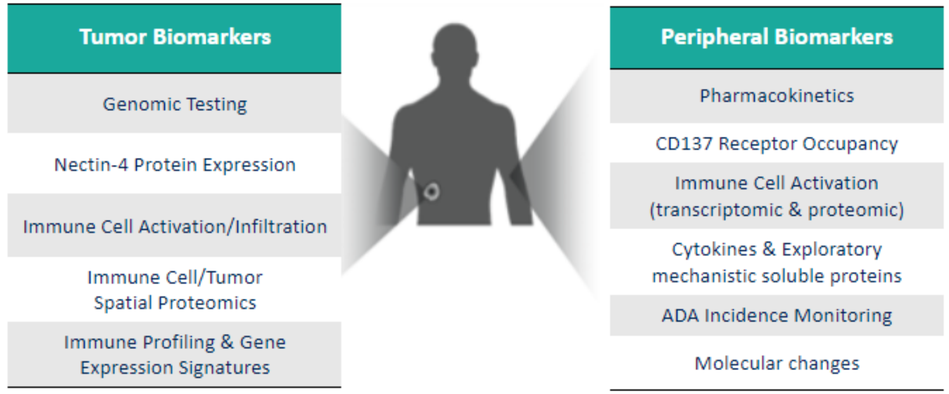




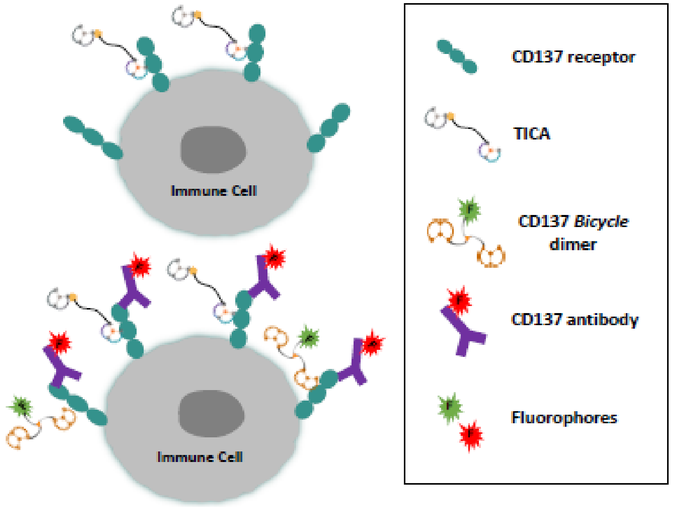
- BT7480 leads to an early increase in cytokine gene expression in tumor
- BT7480 leads to increase in CD8+ cell infiltration, cytotoxic and macrophage cell scores in tumor
- BT7480 induces significant changes in local immune cell populations

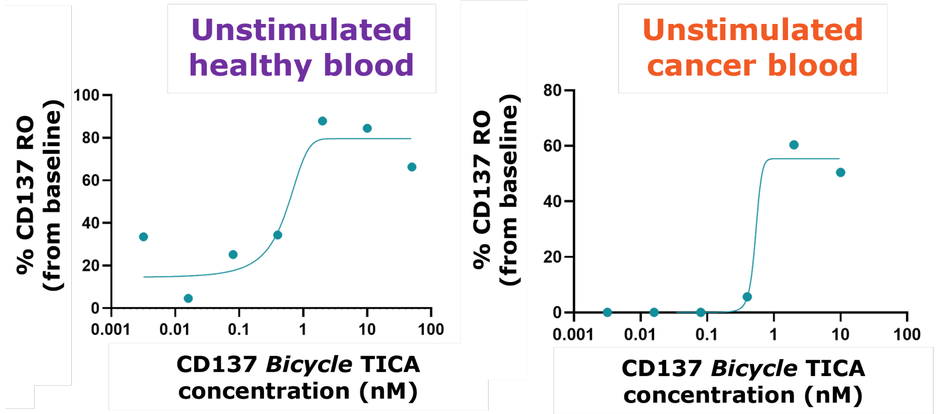
BT7480 now being tested in cancer patients in an innovative biomarker-enabled phase 1 trial

- Phase 1 safety & tolerability study, FIH Nov 2021
- ATD followed by 3+3 escalation, QW IV - tumor response assessed per RECIST every 8 weeks
- Sophisticated biomarker plan to monitor target engagement and immune responses in real time



- Immunophenotyping by flow cytometry on fresh blood drawn after dosing
- Proprietary fluorescent CD137 Bicycle as occupancy probe
- Custom clinical grade RO assay implemented -> guide dose escalation and inform RP2D

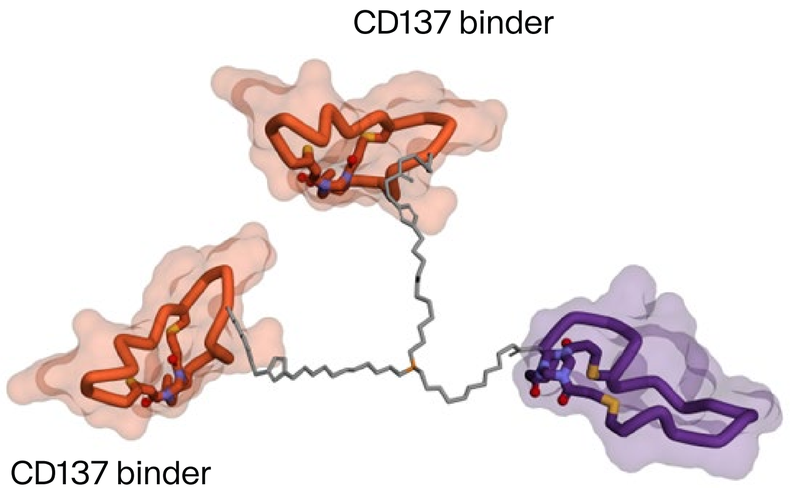




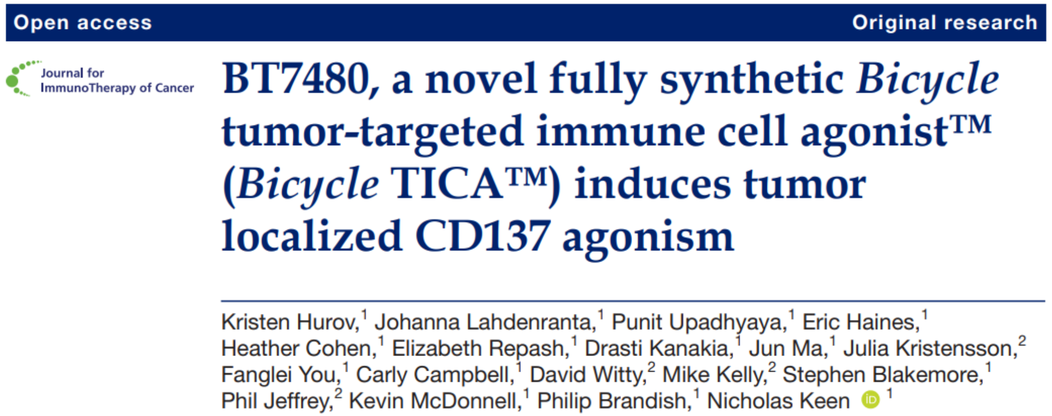
Bicycle®

BT7480 – first chemically synthetic, conditionally active targeted CD137 activator

- Activity of the CD137 agonist arm is dependent on ligation of the Nectin-4 arm, leading to tumor specificity
- Causes complete regressions and anti-tumor activity with only intermittent dosing in syngeneic mouse models
- Causes an early increase in chemotactic cytokine production that precedes an increase in CD8+ T cell infiltration into the tumor
- Is well-tolerated in preclinical safety species
- Entered phase 1 clinical testing in November 2021



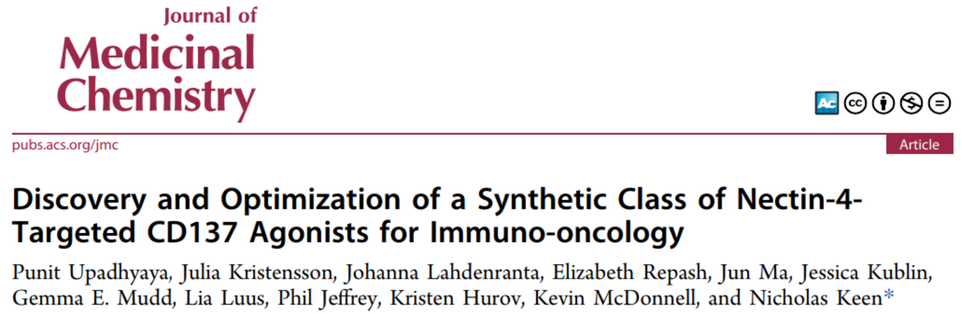
BT7480
MW = 7213.3 Da


Open access Original research

Journal for ImmunoTherapy of Cancer

BT7480, a novel fully synthetic *Bicycle* tumor-targeted immune cell agonist™ (*Bicycle* TICA™) induces tumor localized CD137 agonism

Kristen Hurov,[1] Johanna Lahdenranta,[1] Punit Upadhyaya,[1] Eric Haines,[1] Heather Cohen,[1] Elizabeth Repash,[1] Drasti Kanakia,[1] Jun Ma,[1] Julia Kristensson,[2] Fanglei You,[1] Carly Campbell,[1] David Witty,[2] Mike Kelly,[2] Stephen Blakemore,[1] Phil Jeffrey,[2] Kevin McDonnell,[1] Philip Brandish,[1] Nicholas Keen [1]


Journal of Medicinal Chemistry

pubs.acs.org/jmc Article

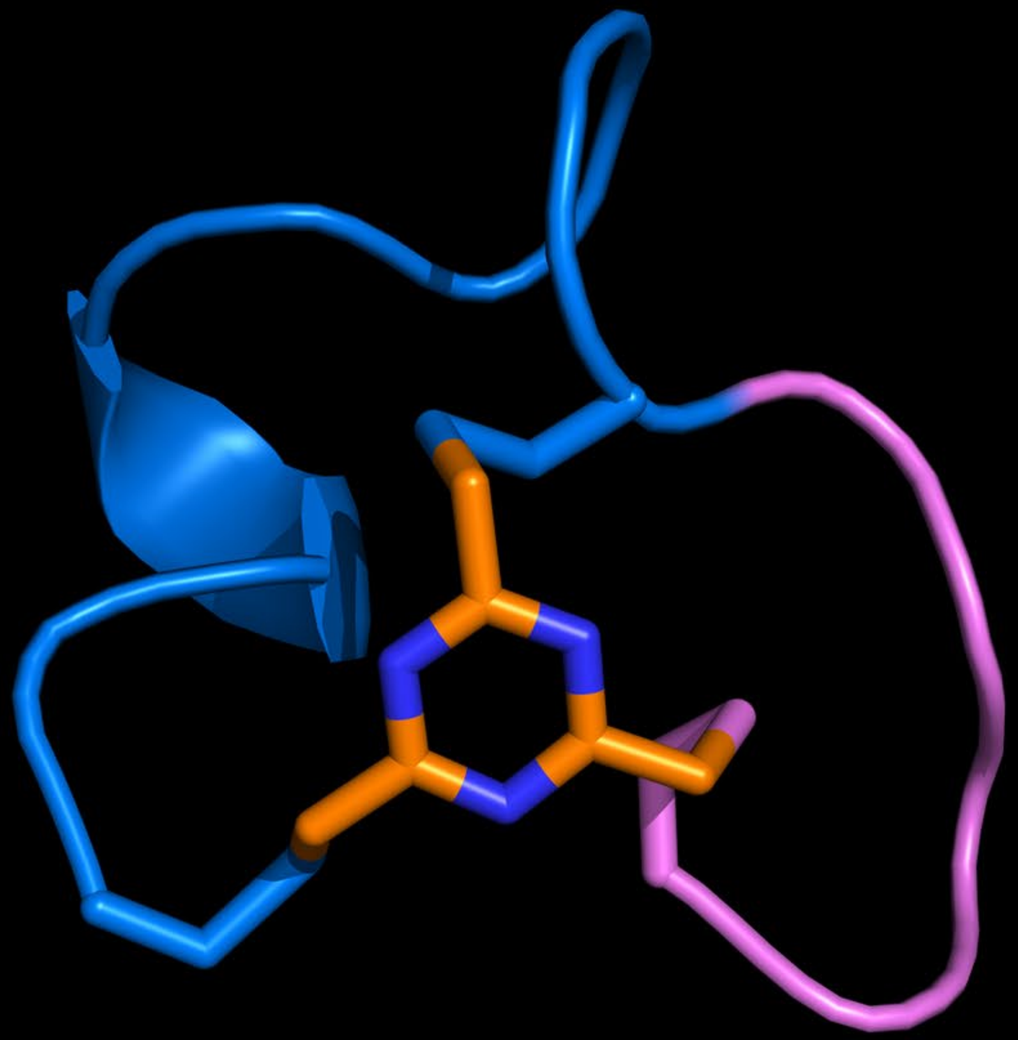
Discovery and Optimization of a Synthetic Class of Nectin-4-Targeted CD137 Agonists for Immuno-oncology

Punit Upadhyaya, Julia Kristensson, Johanna Lahdenranta, Elizabeth Repash, Jun Ma, Jessica Kublin, Gemma E. Mudd, Lia Luus, Phil Jeffrey, Kristen Hurov, Kevin McDonnell, and Nicholas Keen*

Hurov K, Lahdenranta J, et al., 2021, *J Immunother Cancer*, **9**(11):e002883
Upadhyaya, et al., 2022, *J Med Chem*, **65**(14):9858-72

Bicycle® precision-guided NK cell activation

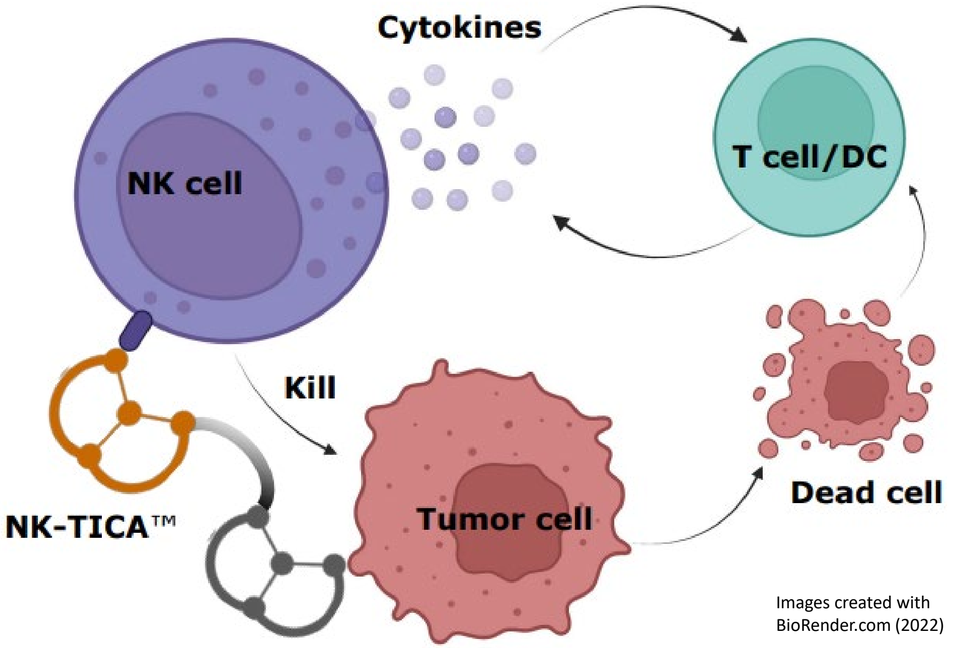
NK cell receptor = NKp46



Bicycle®

Natural killer (NK) cells have emerged as important early drivers of the adaptive anti-tumor immune response

- Traditional understanding – NK cells kill tumor cells one by one through direct cytotoxic mechanisms
- New science: role for NK cells in orchestration of adaptive immunity -> catalysis (Bottcher, 2018)
- NK cell therapy emerging as an important new approach to cancer (Laskowski, 2022)
- NKp46 - an untapped target
  - Activating receptor constitutively expressed on NK cells
  - Not down-regulated in the TME like CD16 or NKG2D
  - Encouraging preclinical data reported (Gauthier, 2019)

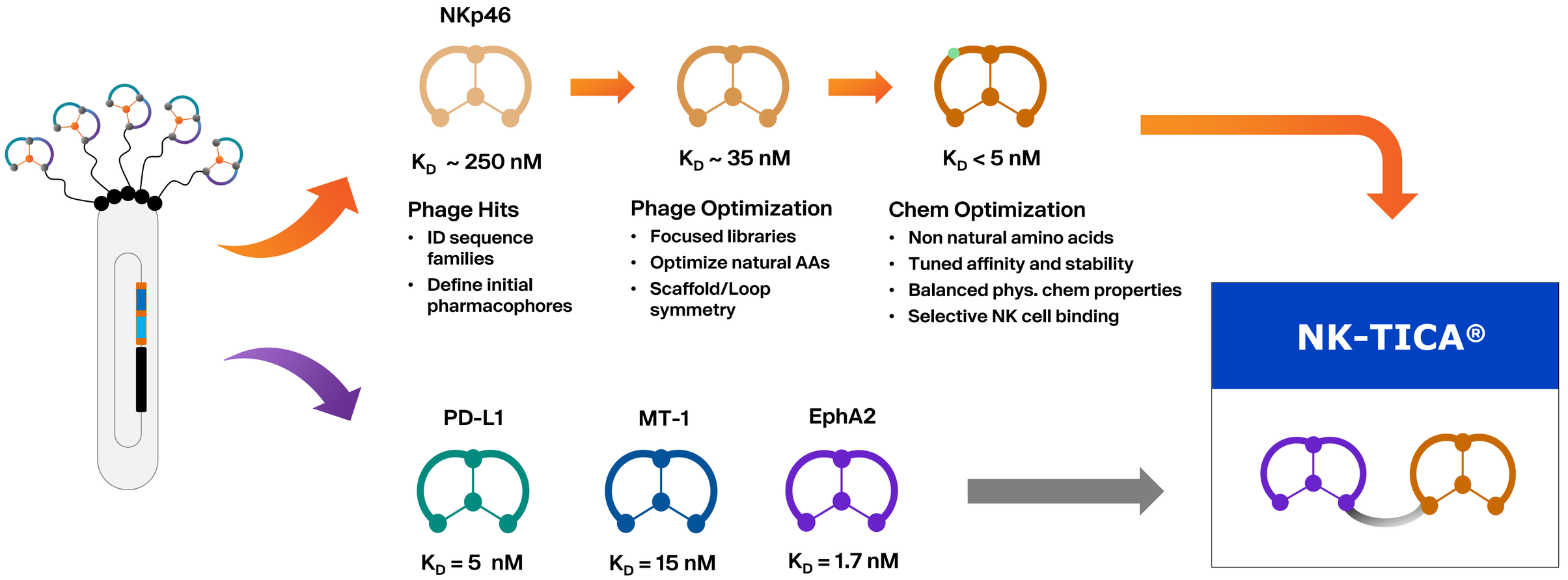


Images created with BioRender.com (2022)

Program hypothesis: Catalysis of adaptive immunity by NK cells has potential to enable tumor rejection and enhance the action of established therapeutics such as targeted toxins and immune checkpoint inhibitors

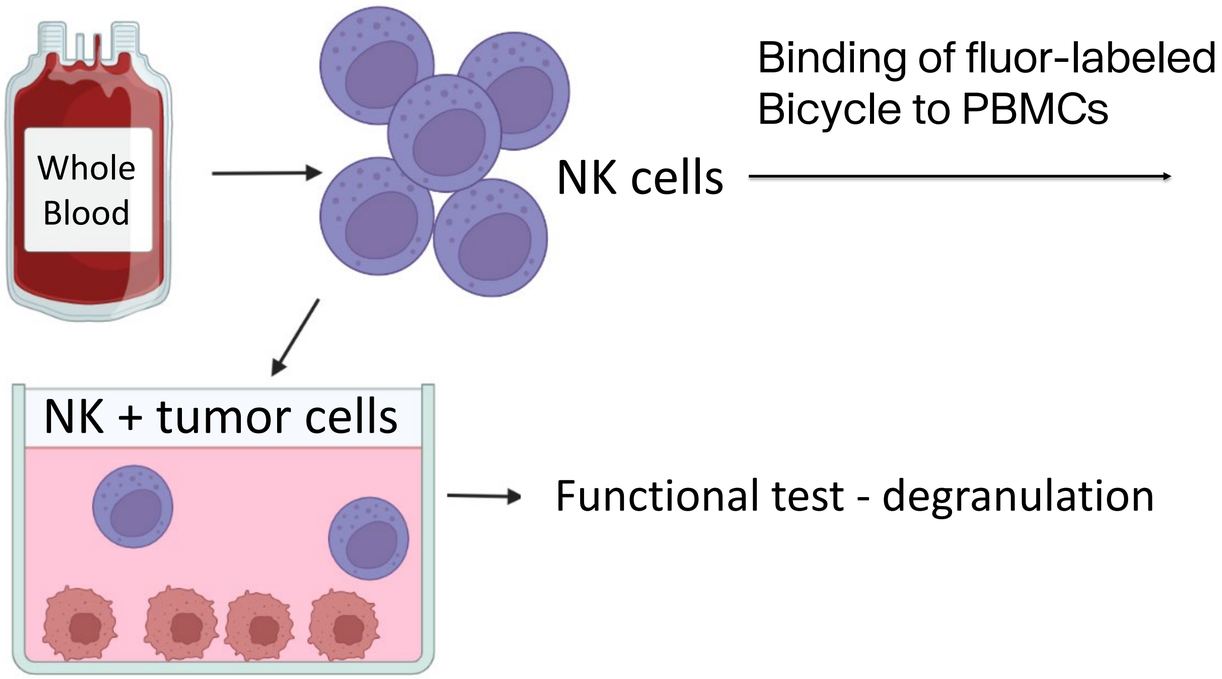
Bald (2020) Nat. Immunol. 21, 835; Barry (2018) Nat. Med. 24, 1178; Bottcher (2018) Cell 172, 1022; Laskowski (2022) Nat. Rev. Cancer 22, 557; Pessino (1998) J. Exp. Med. 188, 953; Gauthier (2019) Cell 177, 1701; Foster (2003) J. Biol. Chem. 278, 46081; Barrow (2019) Front. Immunol. 10, 909

NKp46 *Bicycles*: discovery and optimization by phage display and chemistry



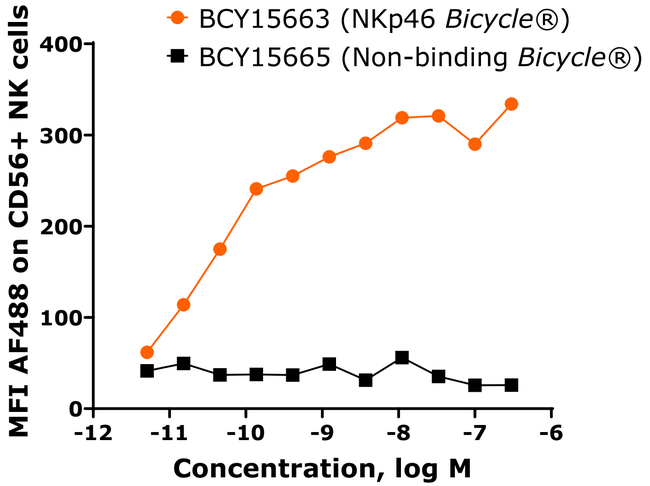
Dufort et al., AACR 2022

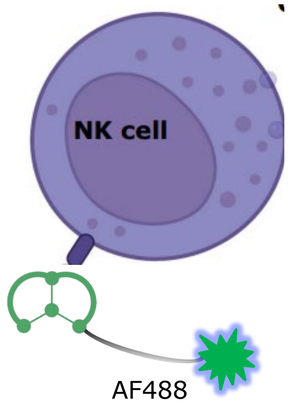
Connecting NKp46 *Bicycles* to a tumor antigen *Bicycle®* quickly led to functionally active molecules

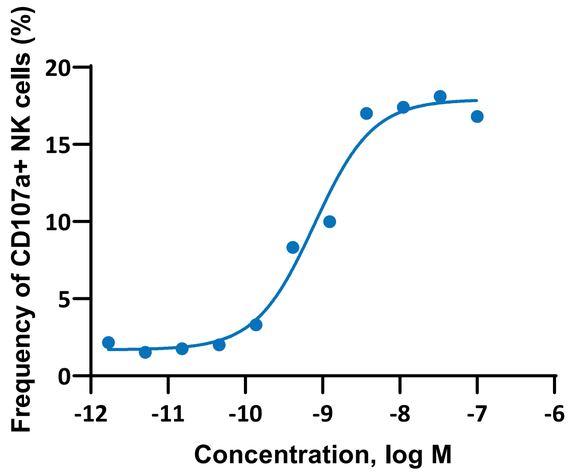


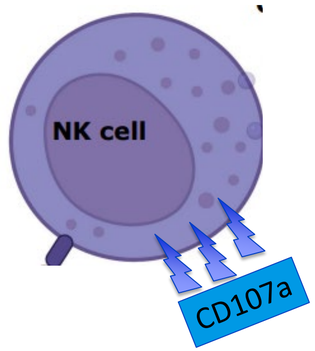
Images created with BioRender.com (2022)

- NK cells isolated from whole blood (negative selection)
- NK cells co-cultured with tumor cell lines in vitro





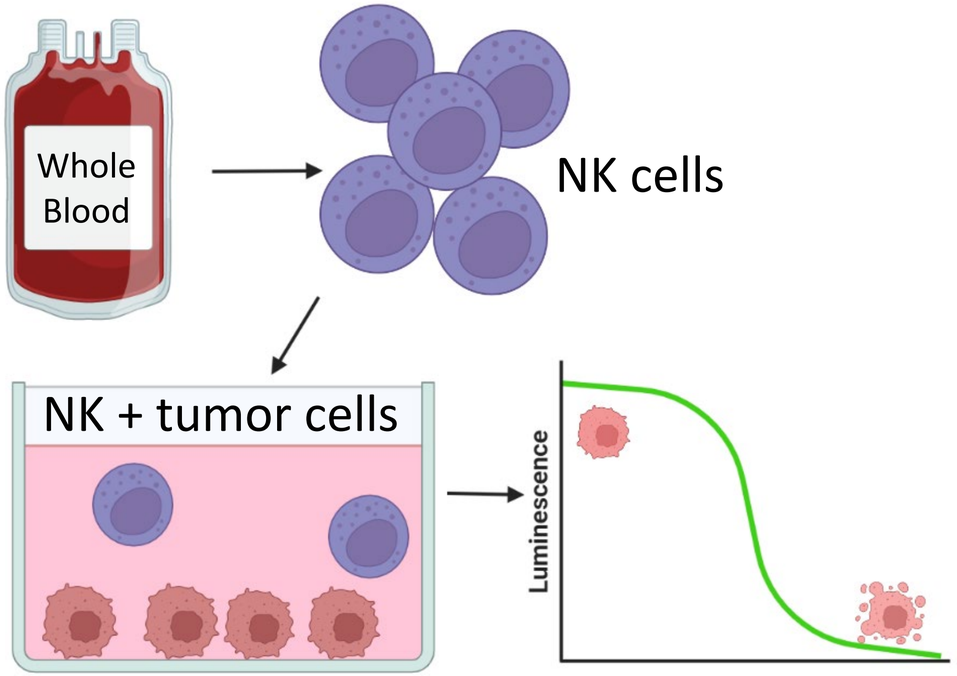




Bicycle®

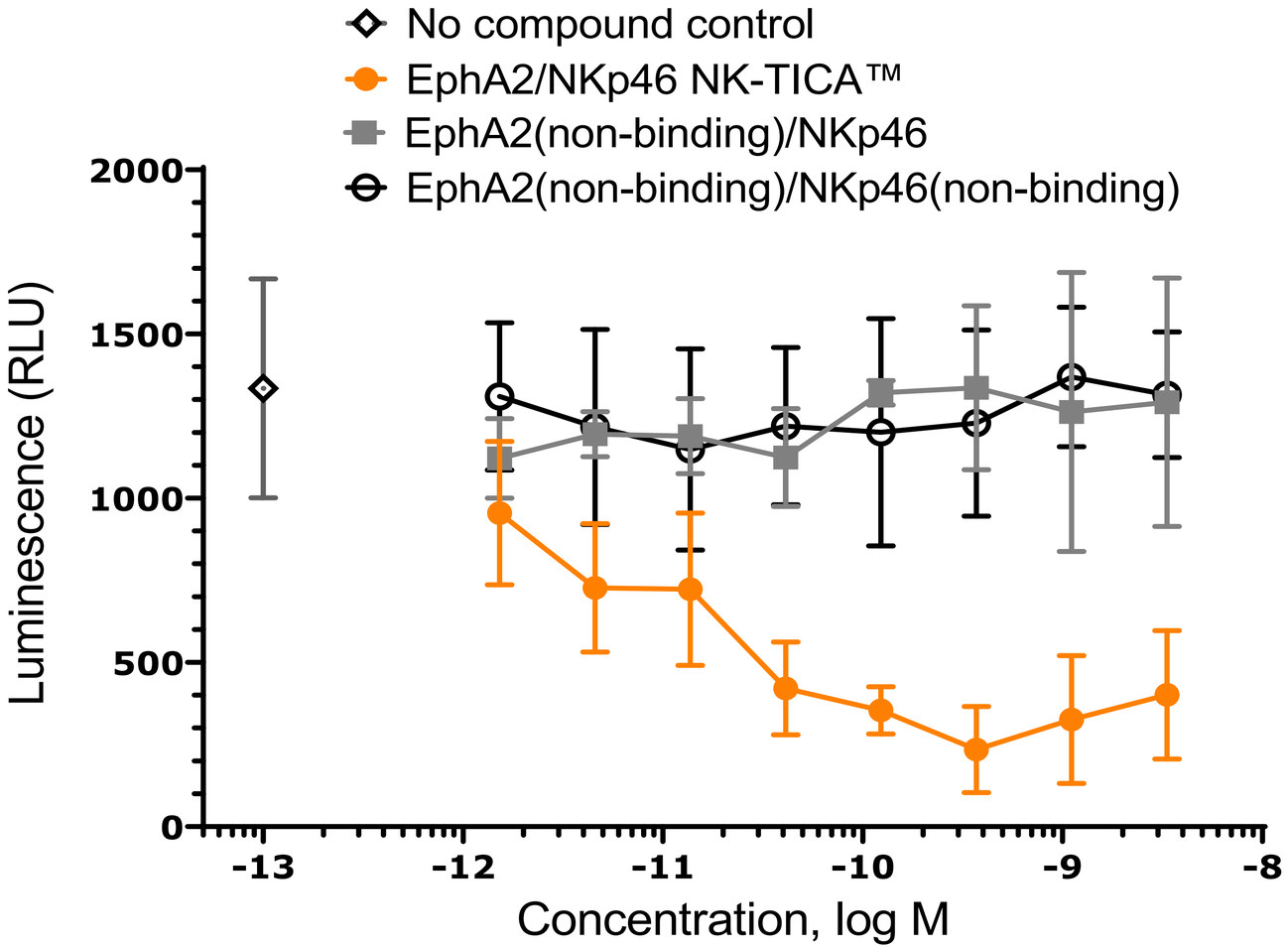
Dufort et al., SITC 2021 & May 2021 BCYC corporate presentation

NKp46/EphA2 *Bicycle*® conjugates enhanced the ability of primary human NK cells to kill target +ve tumor cells



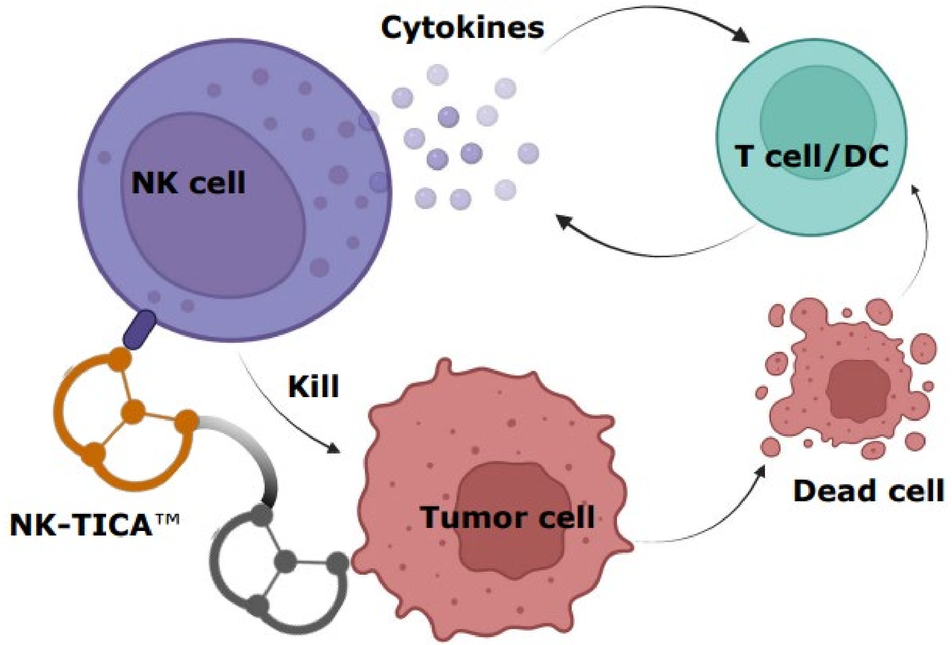
Images created with BioRender.com (2022)

- NK cells are co-cultured with HT1080 cells expressing luciferase in presence of NK-TICA™
- NK cells kill those tumor cells, measured by drop in luminescence

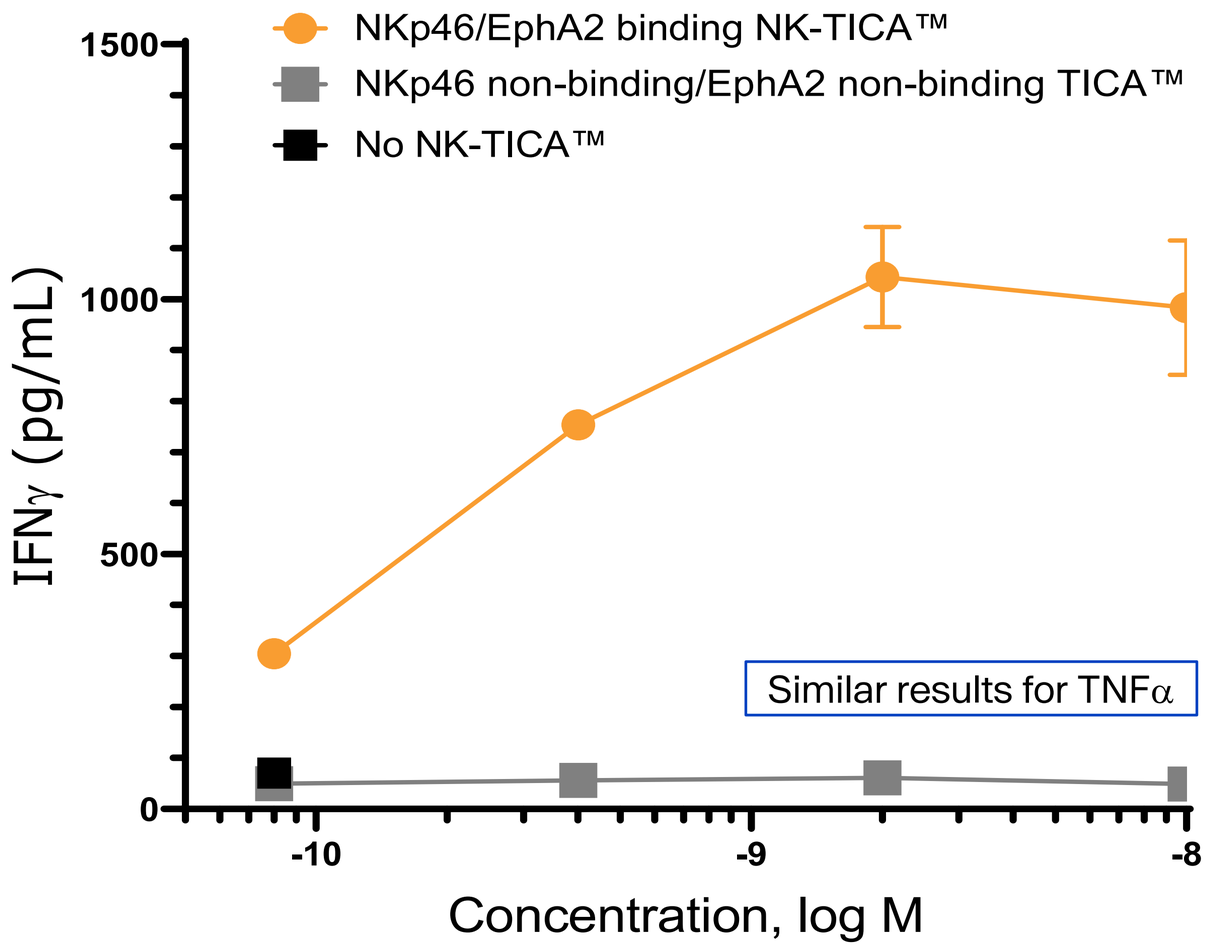


Dufort et al., SITC 2021

EphA2/NKp46 *Bicycles* also cause cytokine production which has the potential to drive adaptive anti-tumor immunity



Tumor target-dependent production of cytokines that can activate APCs is a key design goal

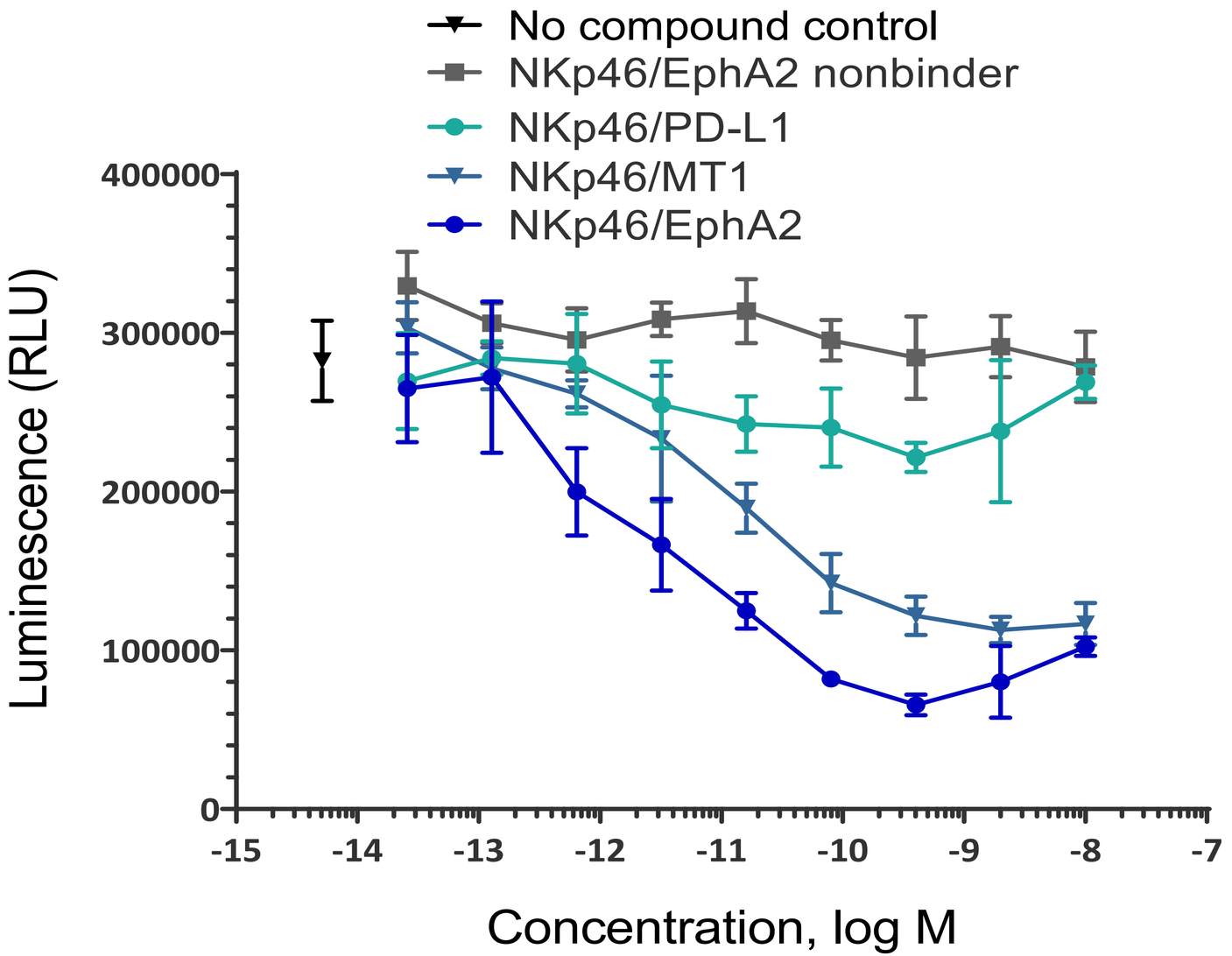


Dufort et al., AACR 2022

NKp46 *Bicycles* work with multiple tumor antigen targets to drive potent tumor cell killing

- HT1080-luc cells in co-culture with primary human NK cells
- HT1080 cells express EphA2, MT-1 and PD-L1

Potential to create NK-TICAs to address multiple solid tumor indications



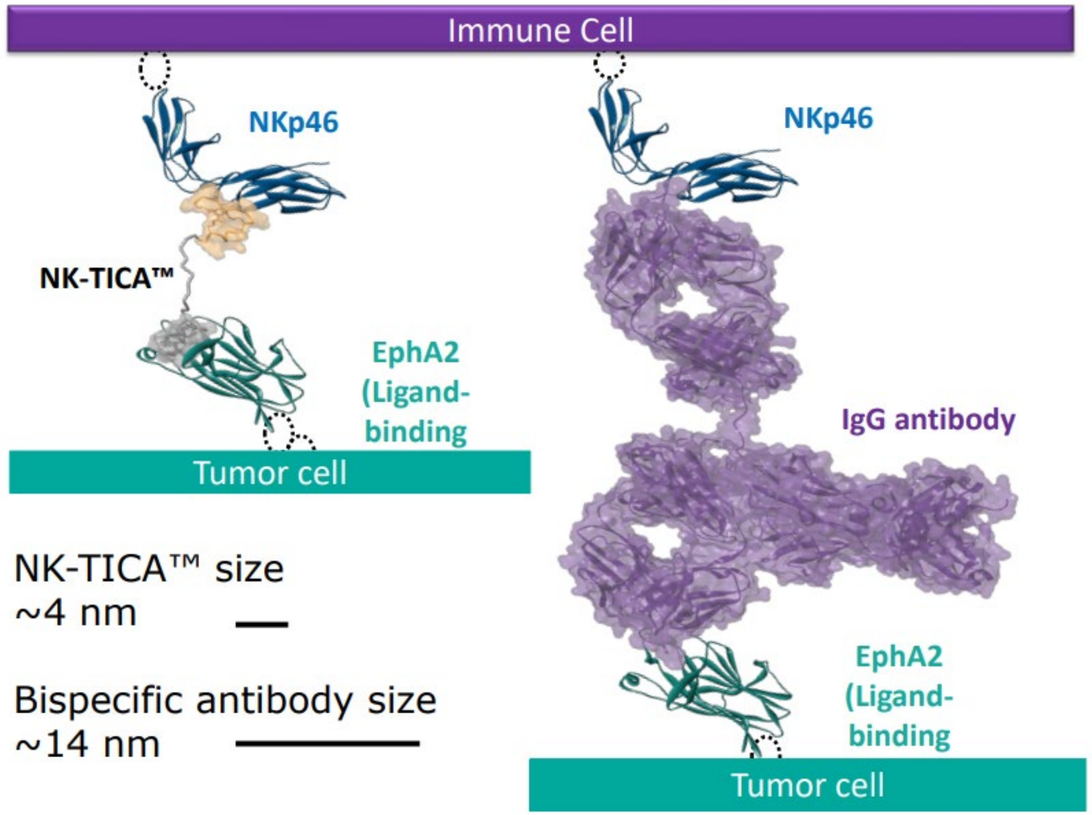
Bicycle®

Dufort et al., AACR 2022

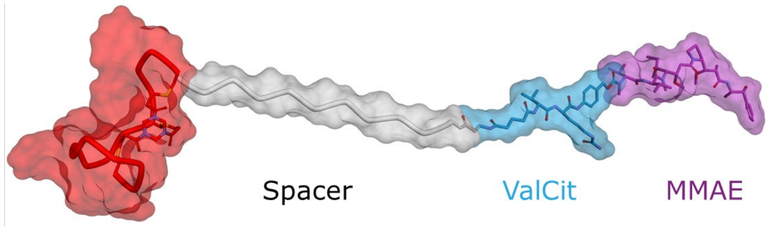
First series of chemically synthetic, conditionally active, targeted NKp46 activators

- Nanomolar biochemical potency
- Sub-nanomolar functional potency
- Directs NK cells to kill target +ve cells
- Drives cytokine secretion
- Activity is tumor antigen-dependent

Potential for activity as a monotherapy and as an adaptor molecule to combine with universal NK cell therapy

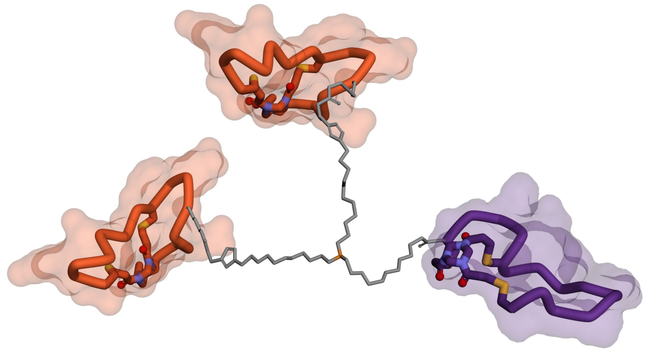


Bicycle Therapeutics – creating versatile new precision-guided medicines with potential to fill major gaps in cancer therapy



Bicycle Toxin Conjugates (BTCs)

- Precision delivery of MMAE - BT8009 & BT5528
- Emerging clinical data



Bicycle Tumor-Targeted Immune Cell Agonists (TICAs)

- Rapid, local and controlled immune agonism
- Pathfinder molecule for CD137 – BT7480 in phase 1
- Pathfinder molecule for NKp46 - preclinical

Thank you

Bicycle®