Azymetric™ Fc-based Therapeutic Modalities Enabling Tumor-Restricted Immune Cell Activation and Engagement

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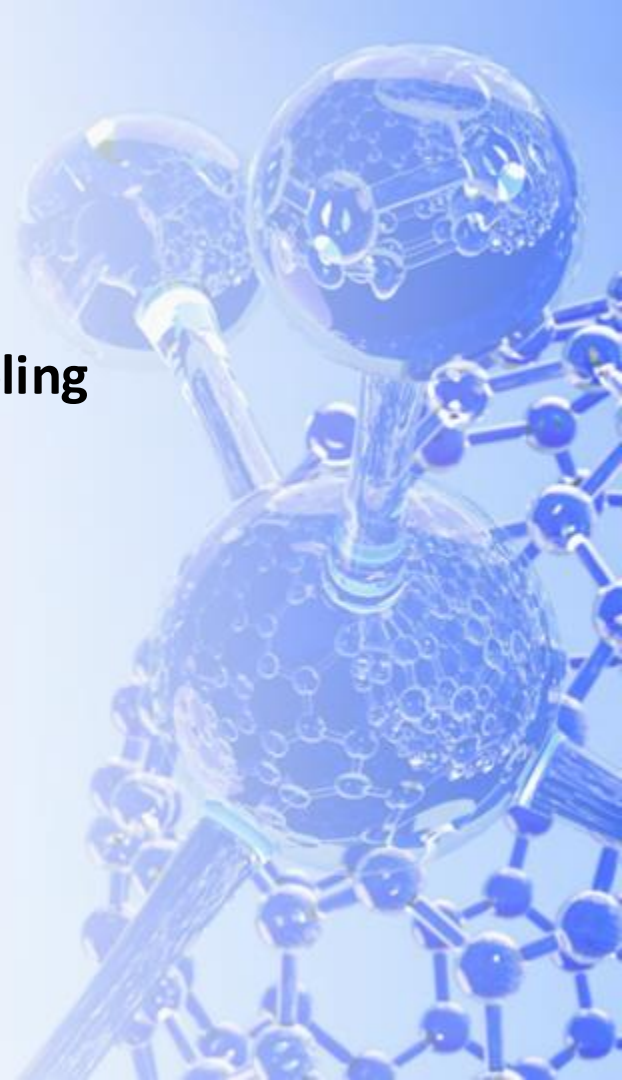
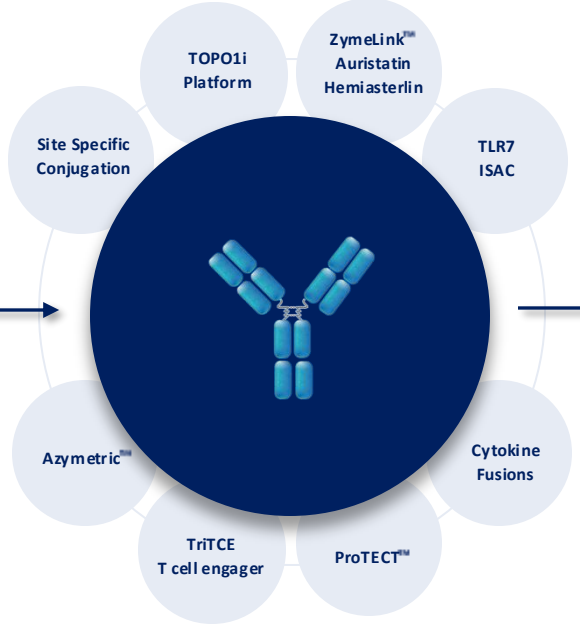


ADC and Multispecific Modalities Driving Zymeworks' Pipeline

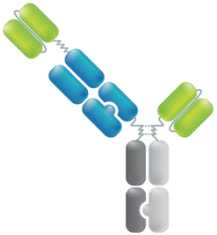
Select Difficult-to-Treat Cancers & Target

Design with Complementary Technology



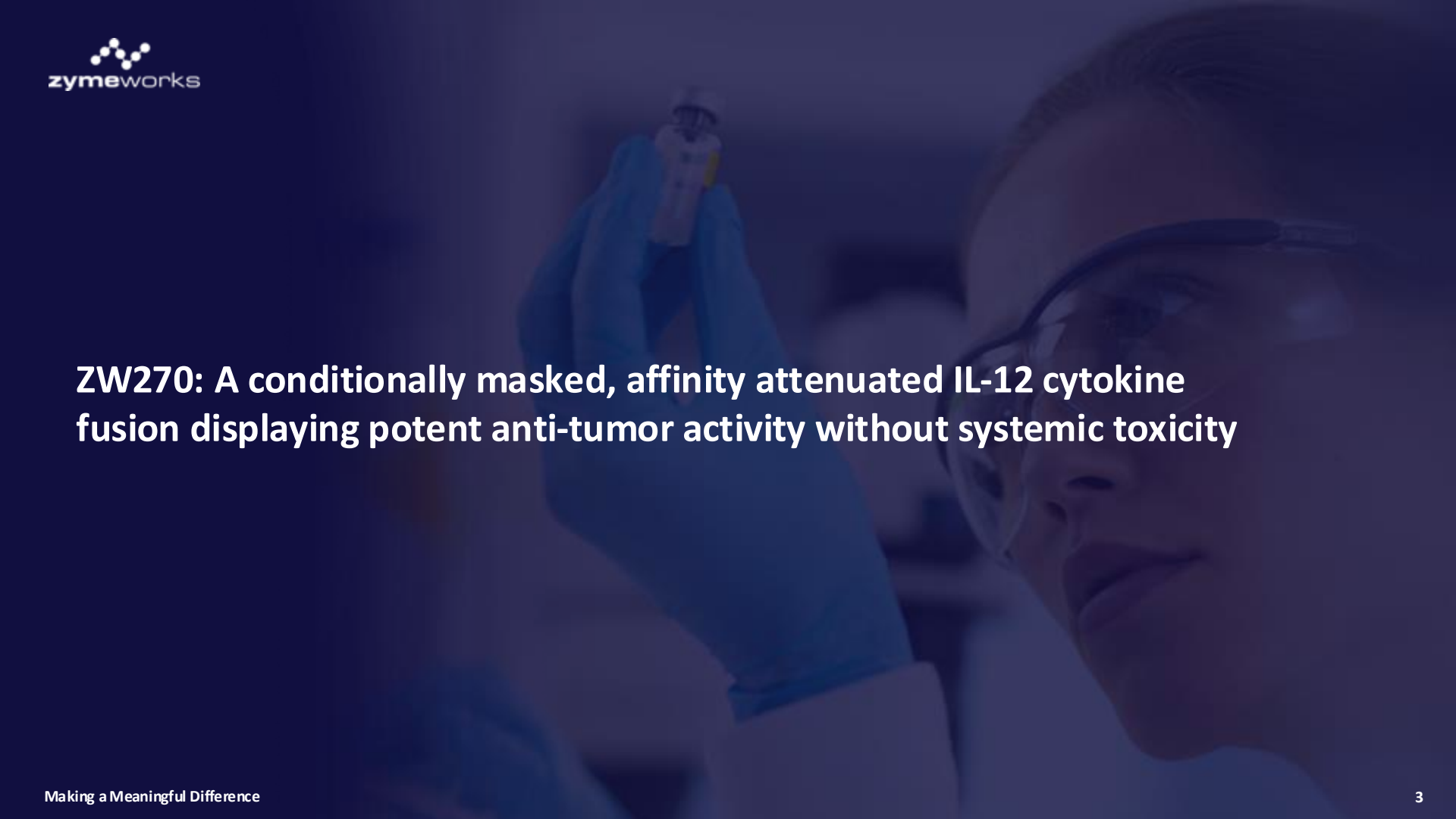
Optionality with Two Foundational Fit-for-Purpose Modalities

Antibody Drug Conjugates
Customization:
- Antibody properties
- Antibody format
- Payload
- DAR



Multispecifics
Customization:
- Multiple MOA in single molecule
- Synergistic biology
- Precision targeting through multivalency

DAR: drug to antibody ratio; ISAC: immune stimulating antibody conjugate; MOA: mechanism of action

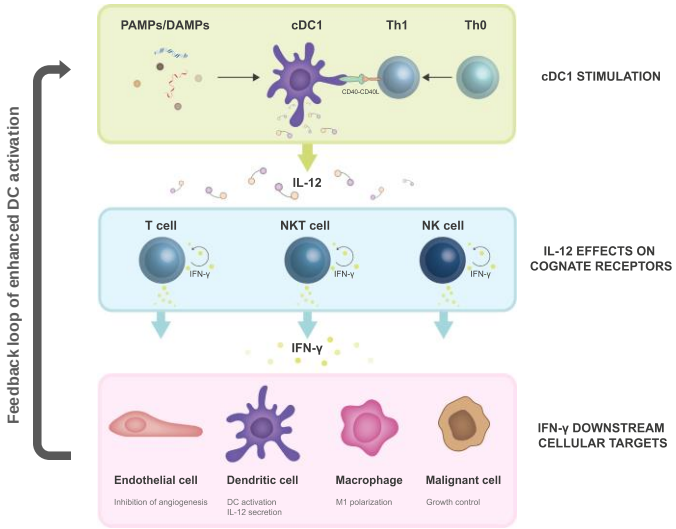


ZW270: A conditionally masked, affinity attenuated IL-12 cytokine fusion displaying potent anti-tumor activity without systemic toxicity

Opportunity for Next Gen IL-12 with Widened Therapeutic Index

- **IL-12** is a highly **potent inducer of anti-tumor immunity**
- Initial clinical responses show ability to turn ‘cold’ tumors ‘hot’, but overall **low responses likely due to toxicity and low TI**
- **Hypothesis:** Widened TI and higher exposure of IL-12 in the TME might increase clinical responses

IL-12 drives innate and adaptive responses



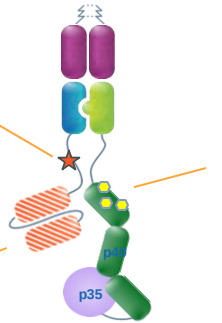
IL-12 Strategies in clinical development

	Development Status
Systemic administration:	
Half life extended IL-12	Most discontinued, lack of TI and clinical responses
Attenuated IL-12 Fc	Discontinued or ongoing, potentially low clinical responses
Conditionally masked IL-12	Ongoing clinical development
Local administration:	
WT IL-12 (recombinant, plasmid)	Discontinued, lack of TI and clinical responses
Targeted IL-12	Ongoing clinical development

Development of a Conditionally Masked IL-12 Cytokine Fusion with Enhanced Therapeutic Window

ZW270: a conditionally masked, affinity attenuated IL-12 Fc

Protease cleavable linker
Proprietary protease cleavage linker with efficient tumor cleavage and slow peripheral release (<1%/day)



Attenuated IL-12 potency
Proprietary mutations to reduce IL-12 potency without affecting scFv masking

Engineered Mask
Anti-IL-12 scFv antibody mask with high affinity to p40, blocking IL-12 activity

Engineering approach: Benchmarking different strategies to optimize TI of IL-12 fusion

1. **WT IL-12 Fc**
   - Wild type (WT) IL-12 potency

2. **Attenuated IL-12 Fc**
   - Reduced IL-12 potency

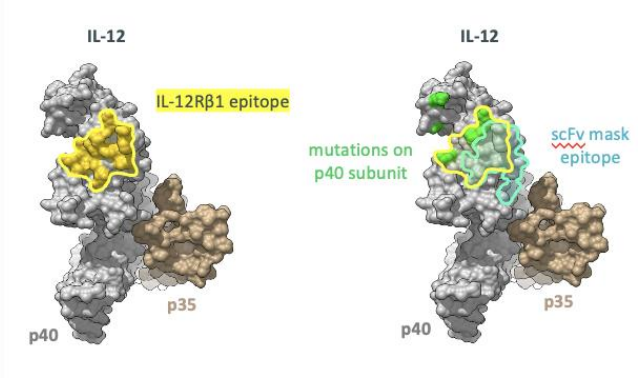
3. **Conditionally masked IL-12 Fc**
   - Cleavable scFv mask
   - Wild type (WT) IL-12

4. **Conditionally masked, attenuated IL-12 Fc**
   - Cleavable scFv mask
   - Reduced IL-12 potency

Combining Masking with Reduced Potency Engineering of IL-12 Leads to Enhanced Masking Window

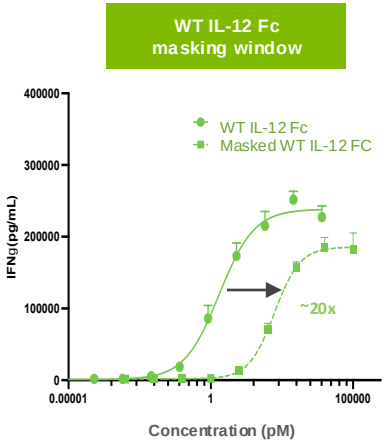
IL-12 was engineered for reduced IL12Rβ1 affinity and IL-12 potency without impacting scFv mask binding

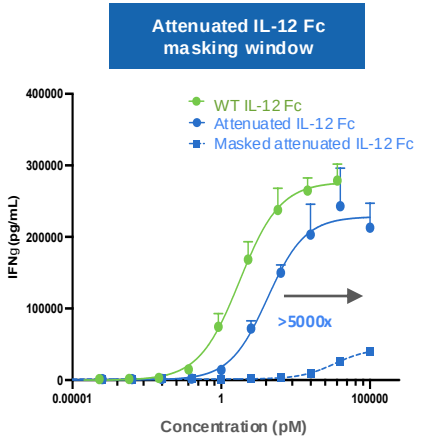
Library of potency attenuated IL-12 variants without impact of scFv mask



Combined masking and attenuation strategies improves masking window in a human *in vitro* T cell activation assay

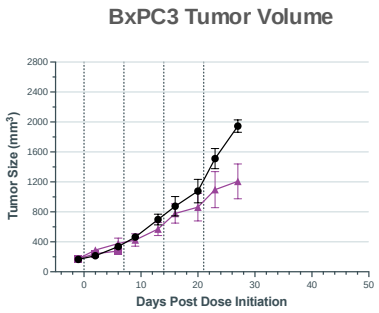
In vitro potency in primary human CD8 T cell assay

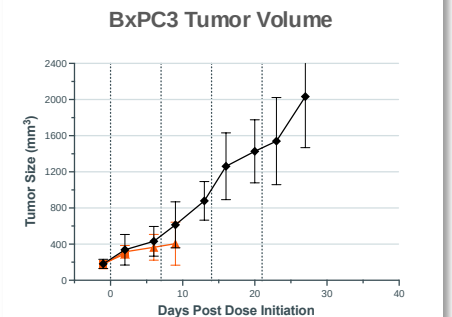


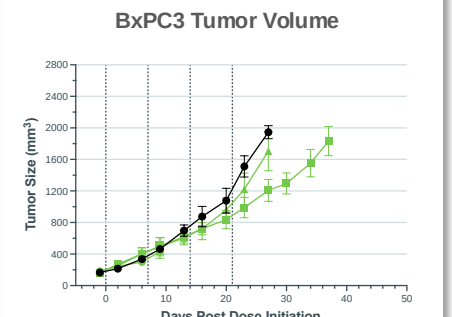


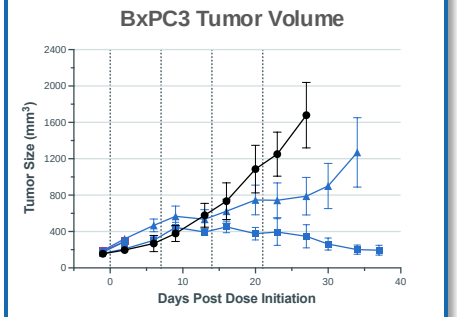
Attenuated, Masked IL-12 Fc (ZW270) Reduces Tumor Growth in a Humanized Mouse Model and is Superior to IL-12 Fc Comparators

	WT IL-12 Fc	Attenuated IL-12 Fc	Masked WT IL-12 Fc	ZW270
Tumor growth inhibition at highest tolerated dose	✗	✗	✗	✓
Maximum tolerated dose (MTD) (<20% loss of mice due to BW loss)	< 0.0008 mg/kg	< 0.008 mg/kg	> 0.01 mg/kg	> 0.1 mg/kg



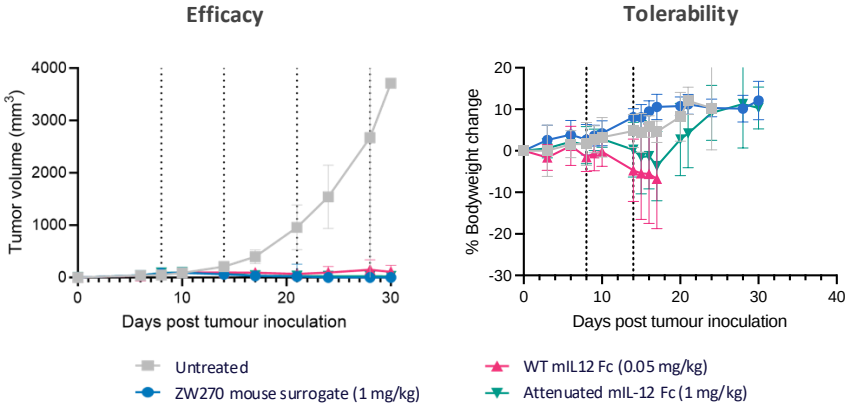






ZW270 Mouse Surrogate Shows Enhanced Therapeutic Window in a Syngeneic Mouse Model

- Development of **mouse surrogate** for attenuated IL-12 Fc and ZW270 (masked, attenuated IL-12 Fc)
- Evaluation of Therapeutic Index (TI) in a **MC38 syngeneic mouse model**



Murine Surrogate	MED (Minimum effective dose)	MTD (Maximum tolerated dose)	TI (Therapeutic Index)
WT mIL-12 Fc*		< 0.05 mg/kg	~10*
Attenuated mIL-12 Fc	0.06 mg/kg	1 mg/kg	~16
Masked, attenuated mIL-12 Fc (ZW270 mouse surrogate)	0.5 mg/kg	32 mg/kg	~32

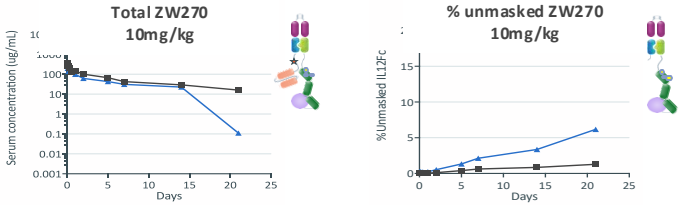
* Gutierrez E et al Med (2023); Koliesnik et al., Cancer Res (2023)

ZW270, a Conditionally Masked IL-12 Cytokine Fusion is well Tolerated in Cynomolgus Monkeys at > 10mg/kg

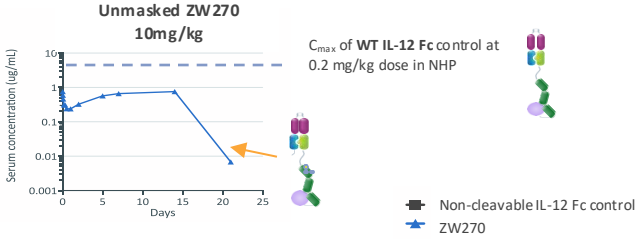
A single dose of attenuated masked IL-12 Fc is well tolerated in cynomolgus monkeys

	WT IL-12 Fc 0.2 mg/kg	ZW270 10 mg/kg
Mortality	**Yes,** (at 0.2 mg/kg, day 21)	No (up to 31.8 mg/kg)
Clinical signs	Watery feces on Day 15; decreased activity on day 8 and 15; thin day 8 and 15; loose non elastic skin day 15	At 10 mg/kg no notable changes At 31.8 mg/kg loose feces on day 15
Body weight, % difference on day 22 compared to pre-dose	**-39.26 %**	-7.56 %
MTD	0.2 mg/kg	> 31.8 mg/kg

ZW270 demonstrates low overall unmasking in cynomolgus monkeys and a slow protease unmasking of < 1%/day



Unmasked IL-12 Fc serum concentration at >10 mg/kg is below C_{max} of unmasked WT IL-12 at MTD in cynomolgus monkeys



ZW270, a Conditional Masked Attenuated IL-12 Fc Cytokine Fusion

Summary

- ZW270 is a novel, masked protease activated IL-12 Fc fusion with attenuated IL-12 potency
- ZW270 has potent anti-tumor activity and a higher therapeutic window than comparator IL-12 fusions
- ZW270 was well tolerated in a single dose non-human primate study up to >30mg/kg
- Our data suggests that combining engineering strategies of affinity attenuation and masking has the potential to widen the therapeutic window of IL-12

ProTECT™: A novel trispecific antibody masking platform with integrated immune modulation displays unique activity and differentiated modes of action

Landscape for Conditionally-Active CD3 Redirection

- **Multiple masking technologies** in development
- Initial promising clinical results, but **low efficacy** remains a potential issue
- All platforms remain susceptible to general limitations of TCEs due to **suppressive TME**

Steric Occlusion

AMUNIX

HARPOON Therapeutics

CentryMed 时迈药业

MAVERICK THERAPEUTICS

Specific Peptide / Antibody Masks

CytomX THERAPEUTICS

MOLECULAR partners

JANUX

Roche

IMMUNORIZON

Conditional Assembly of CD3

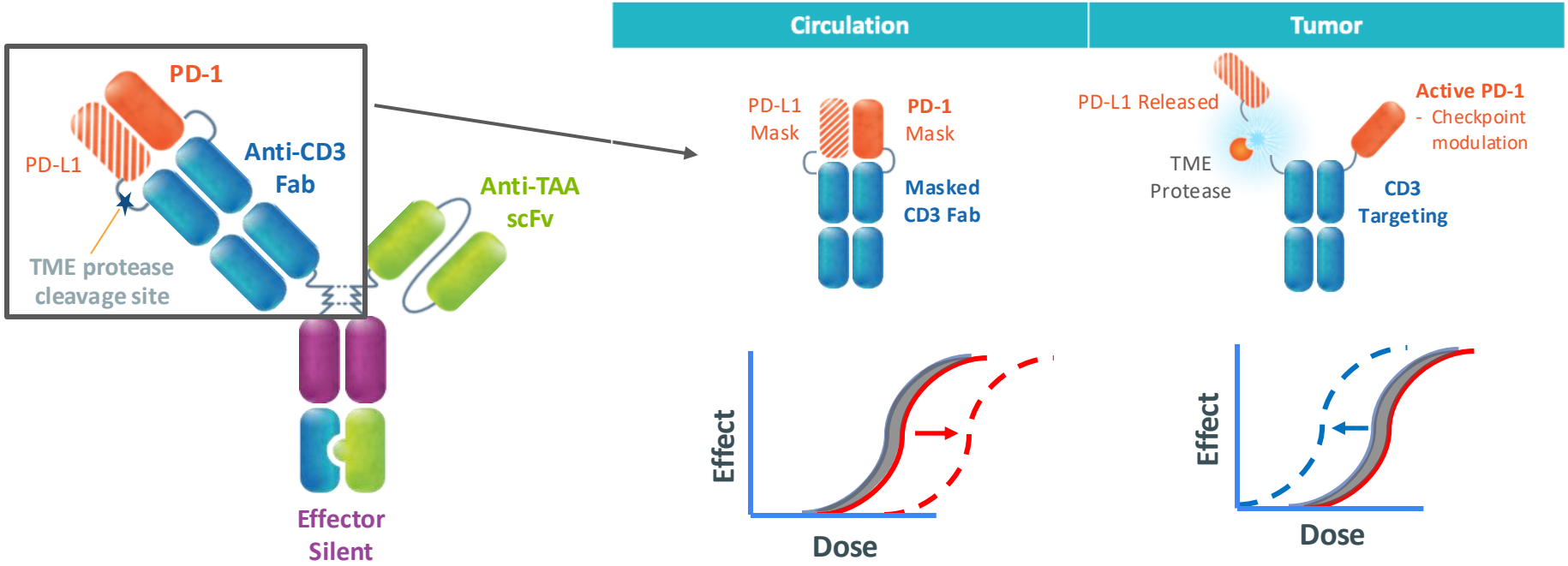
Revitope ONCOLOGY

Soteria BIOTHERAPEUTICS

CHERRY biolabs

ProTECT™ Platform
PROgrammed Tumor Engagement & Checkpoint Targeting

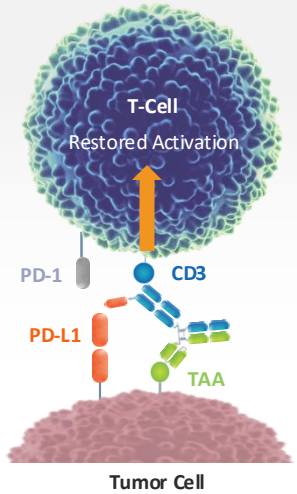
Functional natural PD-1/PD-L1 heterodimer sterically blocks antigen binding



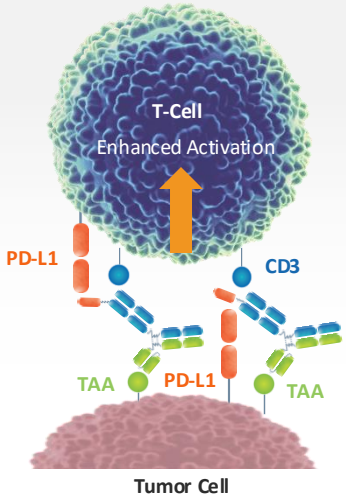
ProTECT™ Has Enhanced Functionality and May Have the Ability to Overcome Limitations of T Cell Engagers in Immunosuppressed Tumors

ProTECT™ trispecific functions by dual MOA and has potential to address limited activity of T Cell Engagers due to PD-1/PD-L1 upregulation in the tumor microenvironment

MOA 1:
Blocking of PD1/L1 inhibition in the synapse restores TCR signal and T cell activation



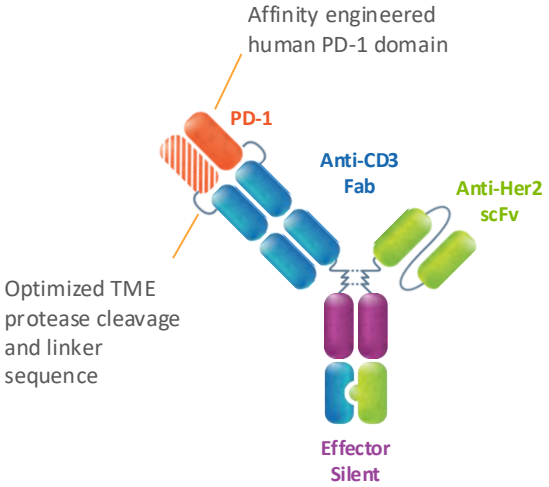
MOA 2:
Enhanced synapse avidity increases activity and T cell activation in tumor microenvironment

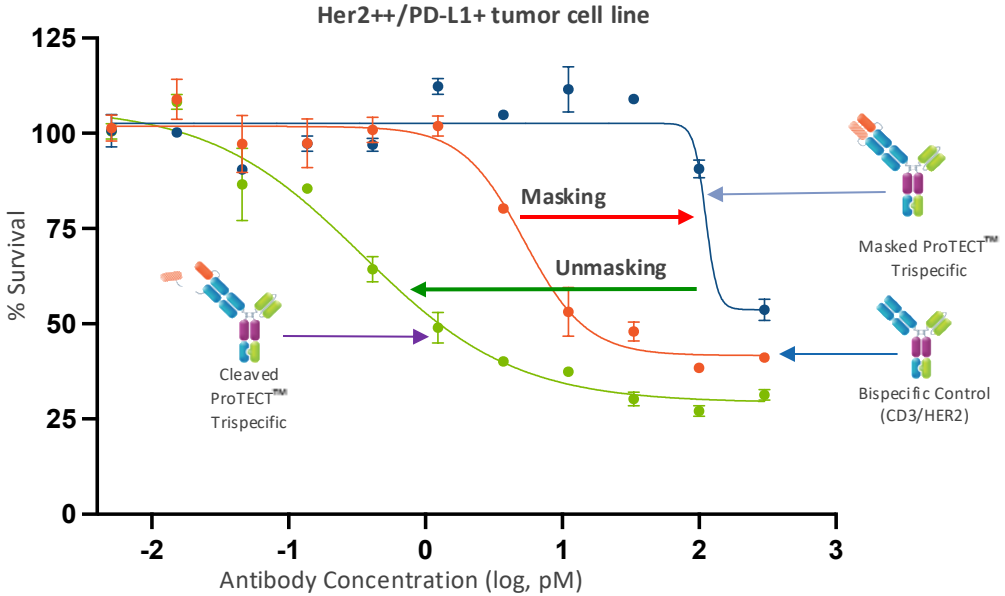


- **PD-1/PD-L1 upregulation** in the tumor microenvironment **is a resistance mechanism** to bispecific T cell engagers
- Enhanced activity of ProTECT™ is driven by **dual MOA** of PD-1/PD-L1 blockade in synapse and increased avidity
- Dual MOA has potential for enhanced activity compared to combination therapy

ProTECT™ Platform
PROgrammed Tumor Engagement & Checkpoint Targeting

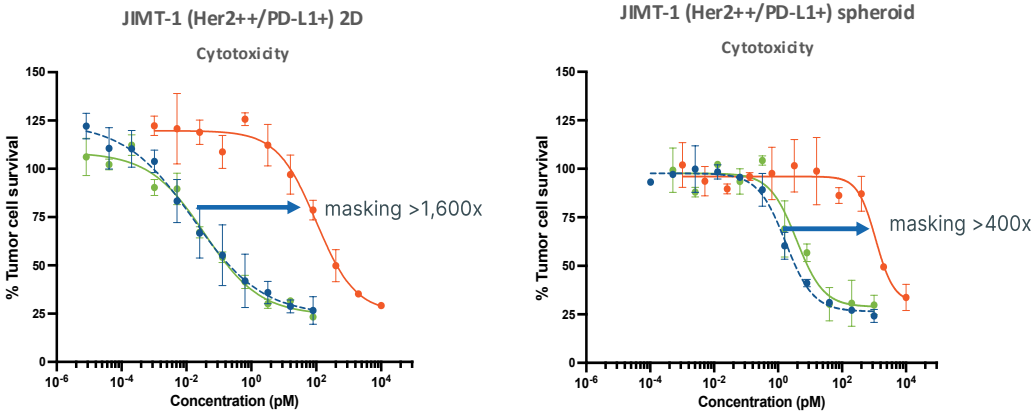
Functional PD-1/PD-L1 heterodimer sterically blocks activity and mediates enhanced *in vitro* cytotoxicity after cleavage





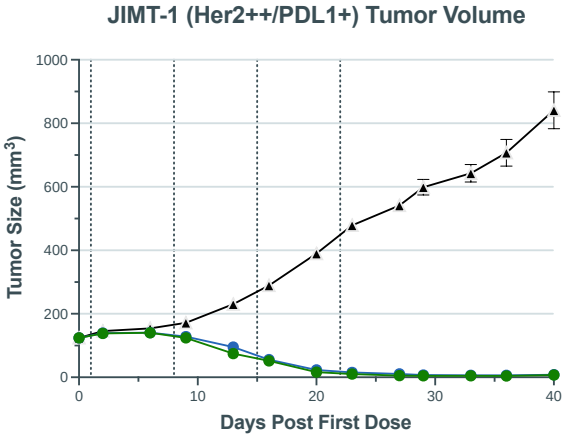
Masked ProTECT™ Mediates Reduced Cytotoxicity While Cleaved ProTECT™ Shows Full Recovery of Cytotoxicity *in vitro* and *in vivo*

Her2xCD3 ProTECT™ has >400x masking window *in vitro* and fully recovers activity after cleavage



- Her2xCD3 ProTECT™ non-masked trispecific
- Her2xCD3 ProTECT™
- Her2xCD3 ProTECT™ non-cleavable

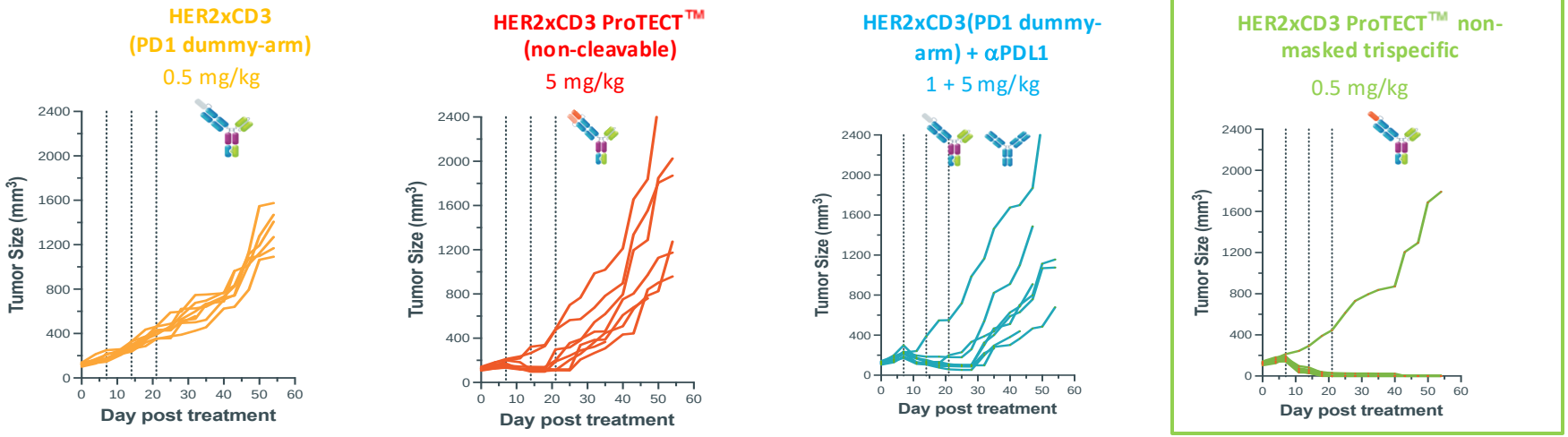
Her2xCD3 ProTECT™ is active in a humanized mouse model



- Her2xCD3 ProTECT™ non-masked trispecific (0.1 mg/kg)
- Her2xCD3 ProTECT™ (0.5 mg/kg)
- Palivizumab (5 mg/kg)

ProTECT™ Induces Complete Individual Tumor Responses in a Humanized Mouse Model and is Superior to Bispecific Anti-PD-L1 Combination

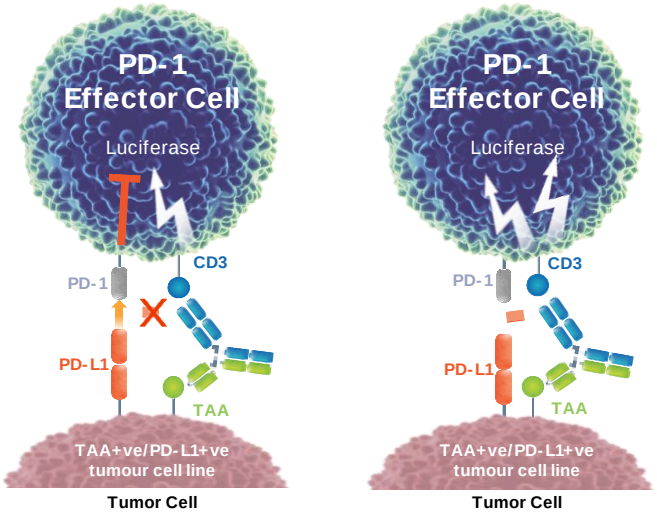
- ProTECT™ trispecific showed individual complete tumor responses
- No complete tumor responses seen for any controls including bispecific Her2xCD3 + Atezo (anti-PD-L1)

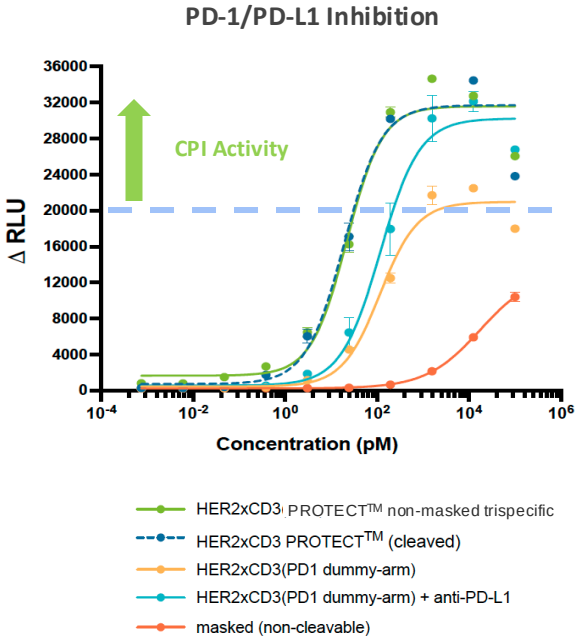


In vivo anti tumor activity of ProTECT™ was assessed in HER2++/PD-L1+ (JIMT-1) tumor model in human PBMC engrafted NOG mice

Protease Activated ProTECT™ Displays Checkpoint Inhibition on PD-L1-Expressing Tumor Cells and is Superior to Combination of Bispecific and Anti-PD-L1

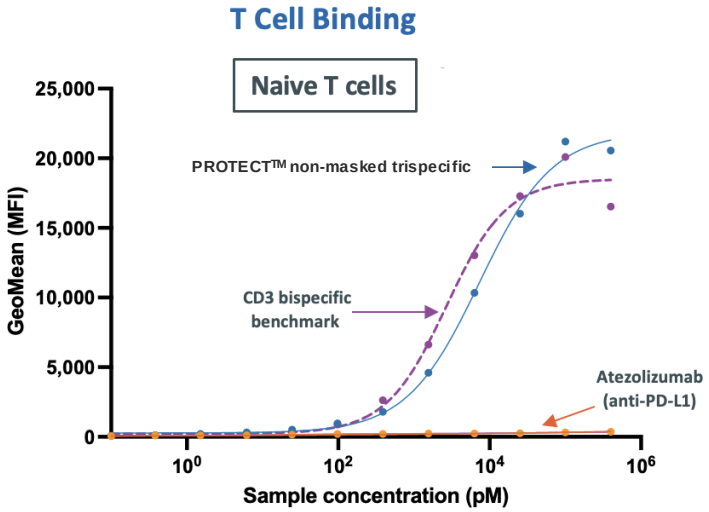
Checkpoint Inhibitor Reporter Gene Assay

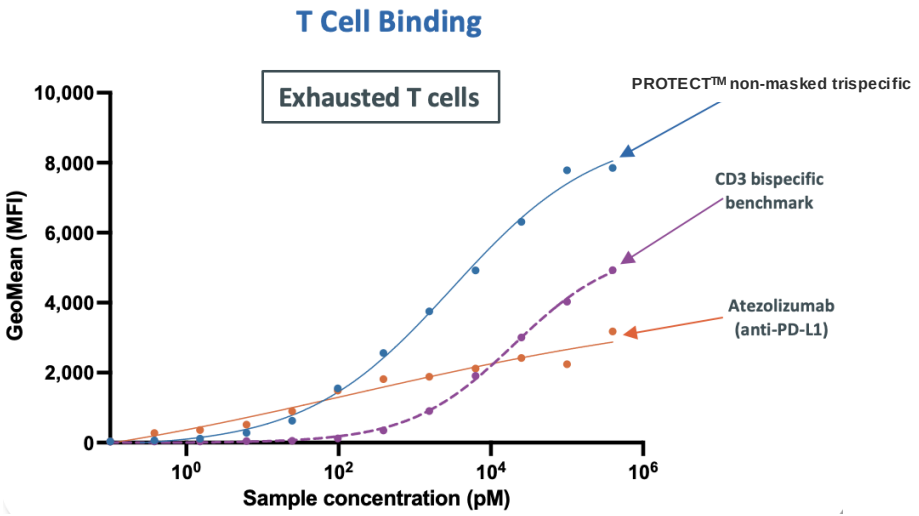




ProTECT™ Trispecific Shows Superior Binding to Exhausted T Cells than Benchmark CD3 Bispecific

- As T cells become activated and exhausted, they express PD-L1 and downregulate CD3*
- ProTECT™ Trispecifics can bind to PD-L1 on exhausted T cells resulting in an avidity advantage

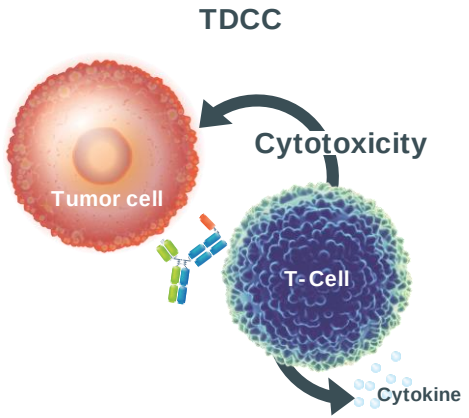


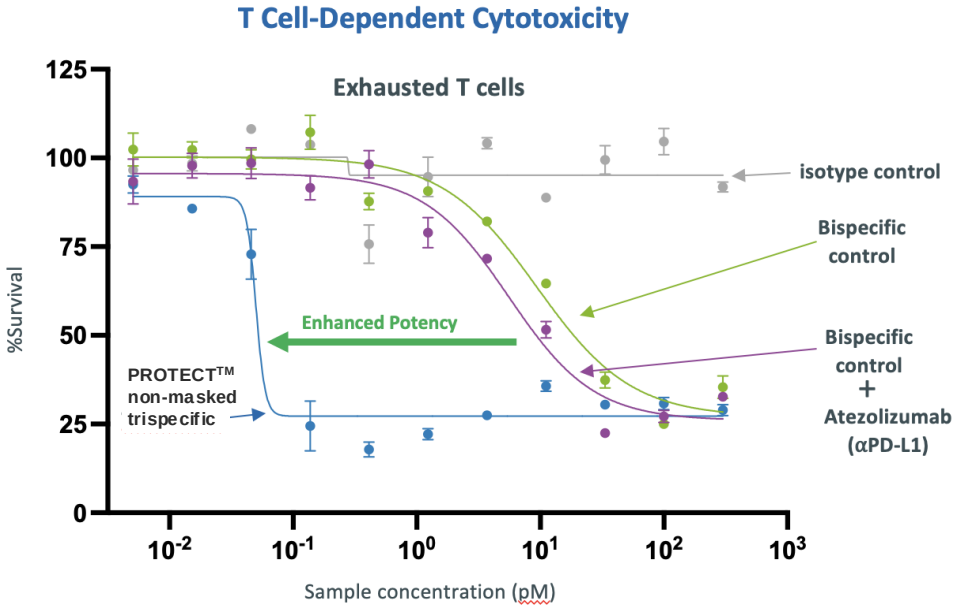


* Lanzavecchia et al., J Exp Med (1997)

ProTECT™ Trispecific Shows Superior *In vitro* Activity to Benchmark with Exhausted T Cells

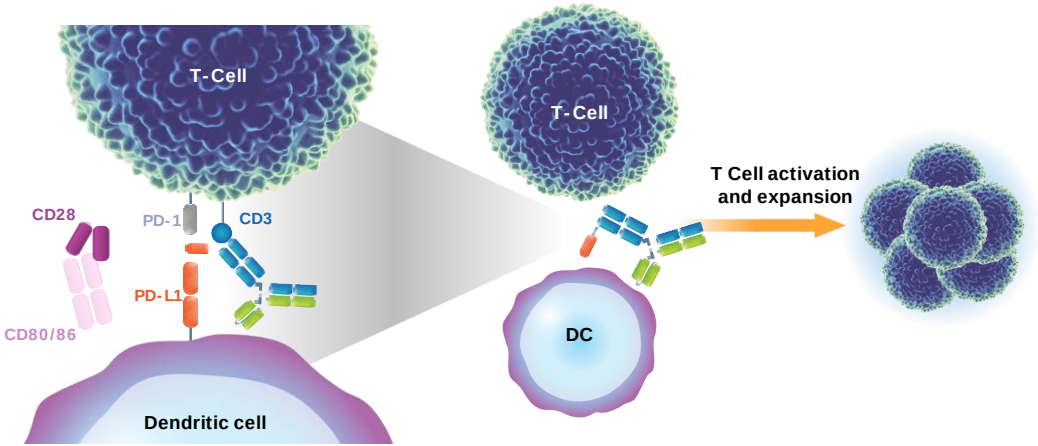
Trispecific shows increased potency compared to bispecific and bispecific + atezolizumab combination when exhausted T cells are used as effector cells



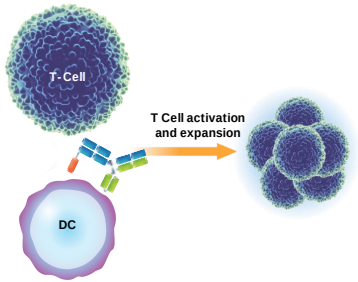


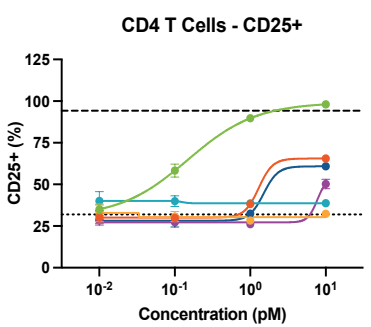
ProTECT™ Trispecific May Have Unique Ability to Induce and Potentiate T cell activation via PD-L1 on APCs in the Tumor Microenvironment

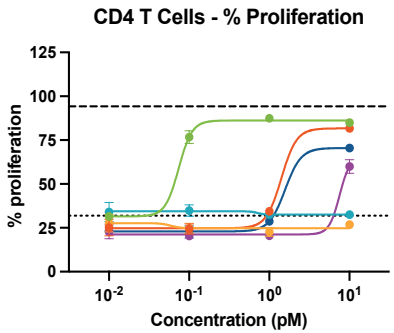
- ProTECT™ trispecific has unique ability to bridge DCs and T cells via PD-L1 and CD3 in the TME
- Potential unique activity of ProTECT™ mediated by crosslinking T cells and PDL1+ myeloid cells in the TME

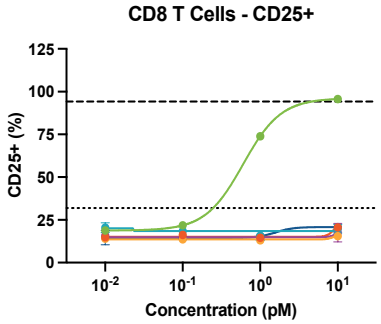


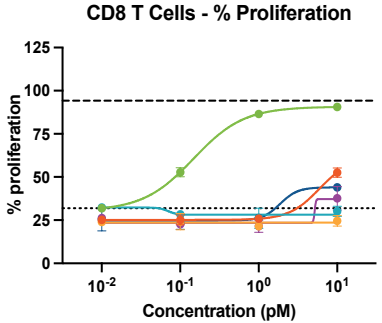
Protease Activated ProTECT™ Trispecific Engages PD-L1-Expressing APCs to Activate T cells









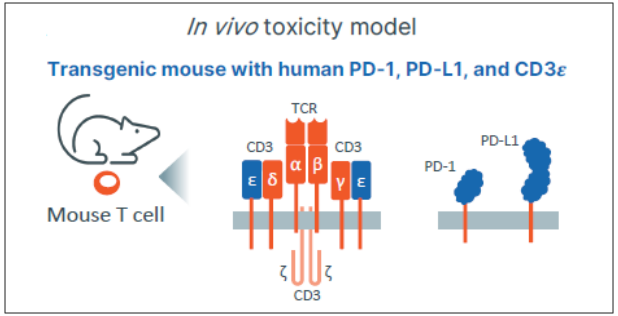


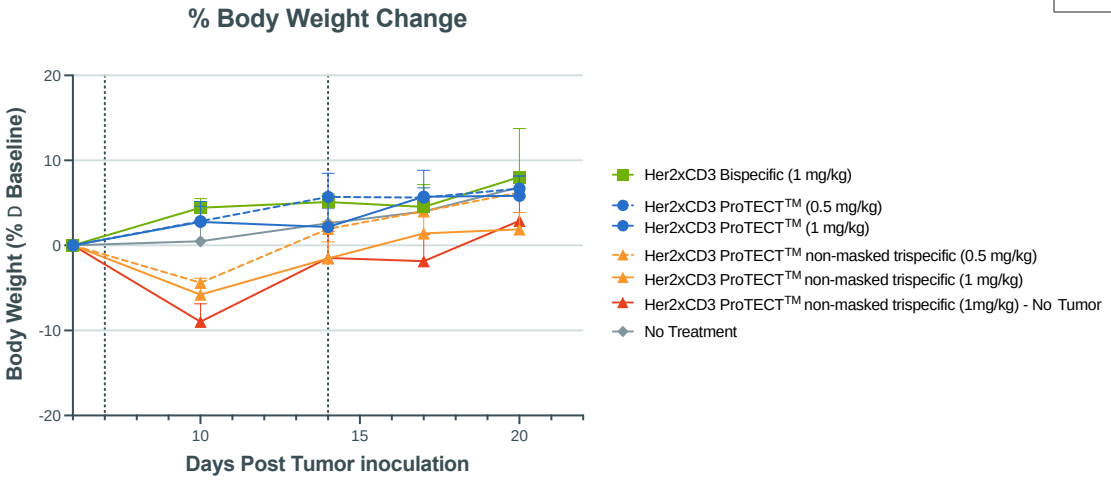
- Her2xCD3 ProTECT™ non-masked trispecific
- Her2xCD3 (PD1 dummy-arm)
- Her2xCD3 (PD1 dummy-arm) + PD1-Fc
- Bispecific control
- Her2xCD3 ProTECT™ (non-cleavable)
- Her2xCD3 ProTECT™

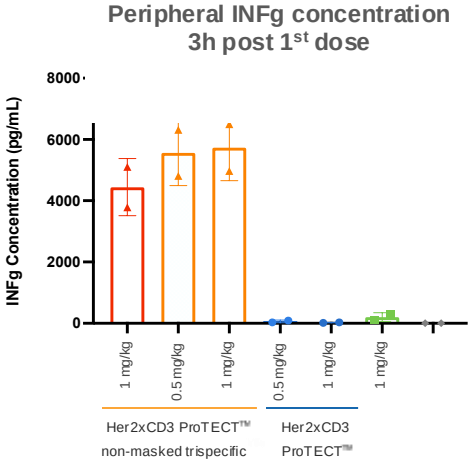
T cells were incubated with autologous PDL1 expressing monocyte derived dendritic cells (moDCs) at a 5:1 ratio for 5 days. % activation and proliferation assessed with CD25+ and CPD dilution respectively by flow cytometry

ProTECT™ is Tolerated in a Humanized Mouse *In vivo* Toxicity Model with Human PD-L1/PD-1 Expression

ProTECT™ is well tolerated in a human PD-L1/PD-1 mouse model, while unmasked trispecific causes peripheral cytokine release







ProTECT™ Platform Summary

- Novel platform that combines tumor-specific masking/unmasking with immune modulation to enhance the therapeutic window for T cell engagers
- ProTECT™ PoC demonstrates effective masking and recovery of activity *in vitro* and *in vivo*
- Demonstrated potent and differentiated activity *in vitro* and *in vivo* and high activity in exhausted T cells
- ProTECT™ PoC demonstrated enhanced functionality and potentially superior activity to combination of TCE plus anti-PD-L1
- Potential for ProTECT™ to be active in suppressive tumor microenvironments and potentially be less susceptible to PD-1/PD-L1 mediated secondary resistance

Acknowledgements…A Global Team Effort

https://www.zymeworks.com/publications/

PROTECT™, A Novel Trispecific Antibody Masking Platform With Integrated Immune Modulation Displays Unique Activity and Differentiated Modes of Action
Anna von Rossum, Genevieve Desjardins, Nichole Escalante, Wingkie Wong, Bryant Harbourne, Janessa Li, Begonia Silva Moreno, Prajwal Raghunatha, Richard Kunze, Madeline Fung, Florian Heinkel, Harsh Pratap, Kevin Haworth, Eric Escobar-Cabrera, Brandon Clavette, Surjit Dixit, Nina Weisser and Thomas Spreter von Kreudenstein

ZW270, A Conditionally Masked IL-12 Cytokine Fusion Protein Displaying Potent Anti-tumour Activity Absent of Systemic Toxicity
Maya C. Poffenberger; Ryan J. Blackler; Kevin G. Haworth; Steven Booth; Shalla Hanson; Jeff R. Proctor; I-Ting Shao; Nichole K. Escalante; Dayananda Siddappa; Joel Smith, Gursev Anmole, Saki Konomura, Nicholas A.J. Dawson; Sifa Arrafi; Desmond Lau; Gerry Rowse; Rupert H. Davies; Thomas Spreter von Kreudenstein

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