# Preclinical Discovery of ARX622, a Site-Specific TLR7-Agonist Antibody-Drug Conjugate

David Mills, PhD
Senior Director, Preclinical Science
November 14-16; PEGS Europe 2023

AMBRX

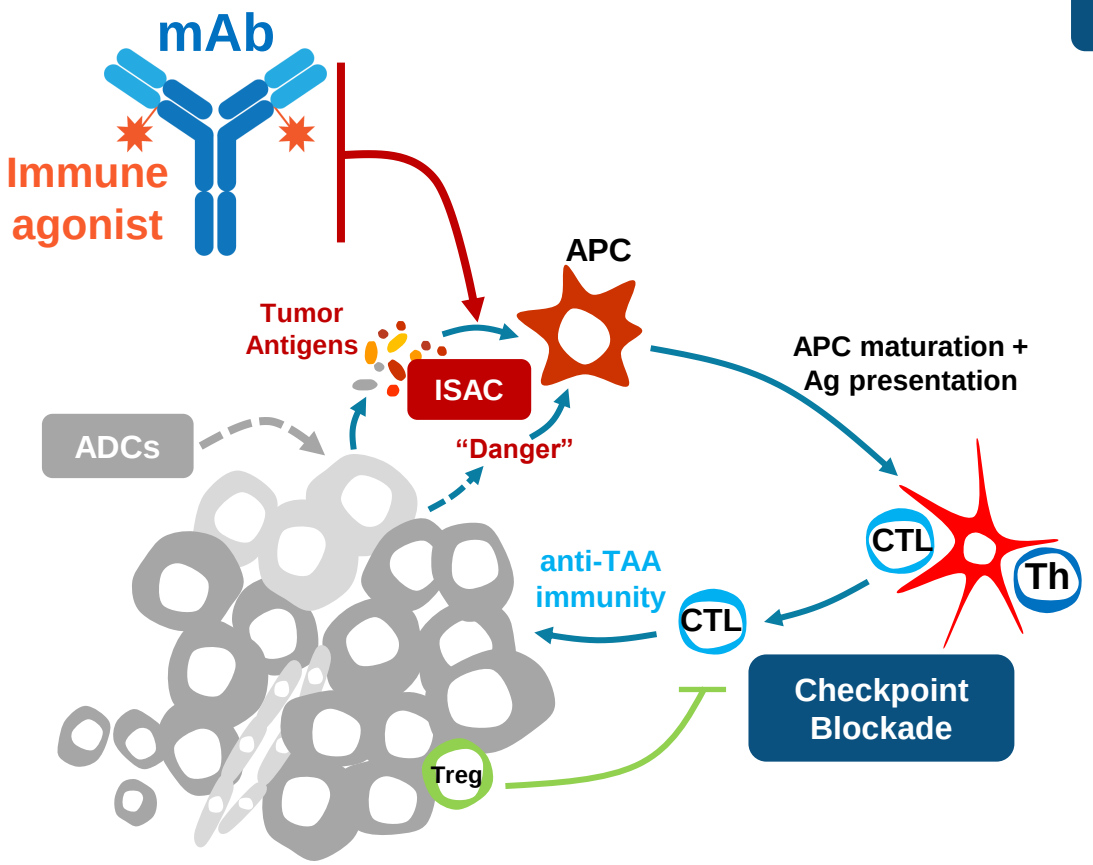
# Forward-Looking Statements

Certain statements contained in this presentation, other than statements of historical fact, are "forward-looking" statements, within the meaning of the Private Securities Litigation Reform Act of 1995. Forward-looking statements can be identified by the use of words such as “believes,” “expects,” “hopes,” “may,” “will,” “plan,” “intends,” “estimates,” “could,” “should,” “would,” “continue,” “seeks,” “pro forma,” or “anticipates,” or other similar words (including their use in the negative), or by discussions of future matters. These forward-looking statements include, without limitation, statements regarding the timing, progress and results of preclinical studies and clinical trials for our product candidates; our product development plans and strategies; plans and expectations with respect to regulatory filings and approvals; the potential benefits and market opportunity for our product candidates and technologies; expectations regarding future events under collaboration and licensing agreements, as well as our plans and strategies for entering into further collaboration and licensing agreements; and expectations regarding our future financial position and results of operations.

Forward-looking statements are subject to risks and uncertainties that could cause actual results to differ materially, and historical results should not be considered as an indication of future performance. These risks and uncertainties include, among others, risks inherent in the development and regulatory approval process for novel therapeutics; the fact that future preclinical and clinical results may not be consistent with prior results; potential delays in development timelines, including delays in clinical trials; the potential impact of the COVID-19 pandemic; our reliance on third parties for development and manufacturing activities; changes in competitive products or in the standard of care; the risk of early termination of collaboration agreements; the risk that our proprietary rights may be insufficient to protect our product candidates or that we could infringe the proprietary rights of others; the fact that we will need additional capital and such capital may not be available on acceptable terms or at all; and changes in laws and regulations. Other factors that may cause our actual results to differ from current expectations are discussed in our filings with the U S Securities and Exchange Commission, including the section titled “Risk Factors” contained therein.

These forward-looking statements are based on information available to, and expectations of, Ambrx as of the date of this presentation. Ambrx disclaims any obligation to update these forward-looking statements, except as may be required by law.

# Immune-Stimulatory ADCs (ISACs) Provide Targeted "Danger" Signals and Complement Multiple Established Treatment Modalities




## The "Danger Hypothesis:" non-self + danger = immunity

"I would suggest that the criteria have to do with what is **dangerous** rather than what is 'self'."

*Polly Matzinger*

*Annu. Rev. Immunol 1994. 12:991-1045*

## ISACs: Multiple Mechanistic Advantages

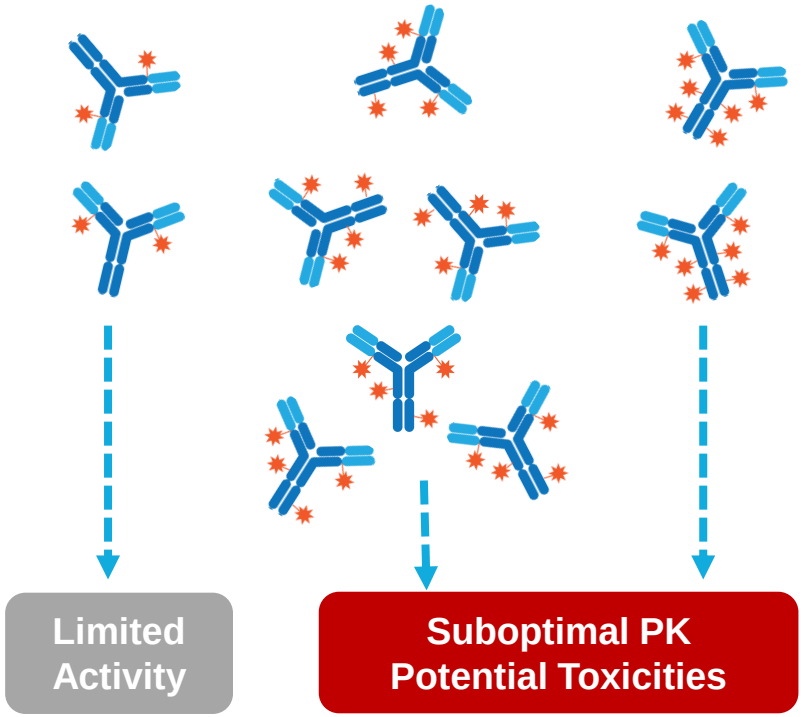
- **Tumor-specific activity with systemic administration**
- **Broad "danger" signal induction (TNF/IL-6, IFNs, chemokines)**
- **APC maturation + polyclonal Ag presentation**
- **Complementary MOA to: ADCs, checkpoint blockade, others**

**New ISAC field can learn from decades of ADC experience**

# DAR Heterogeneity and Instability Can Limit the Potential of Traditional ADCs With Stochastic Conjugation

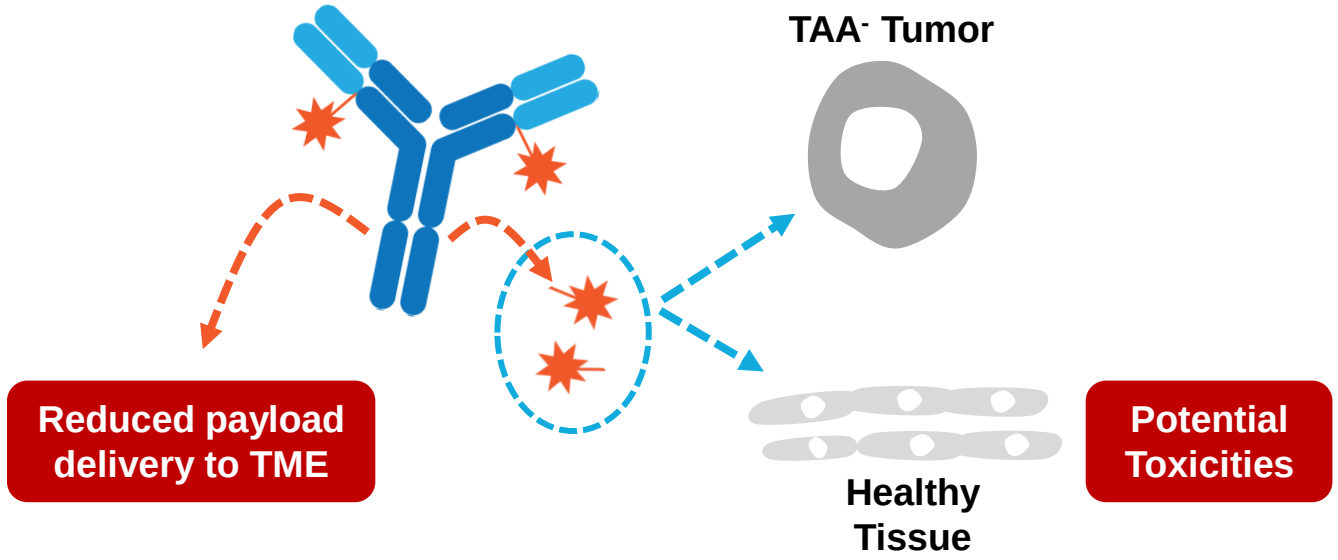
## Heterogeneous DAR Mixture:

Low DAR → High DAR




## Labile Linker/Conjugations:

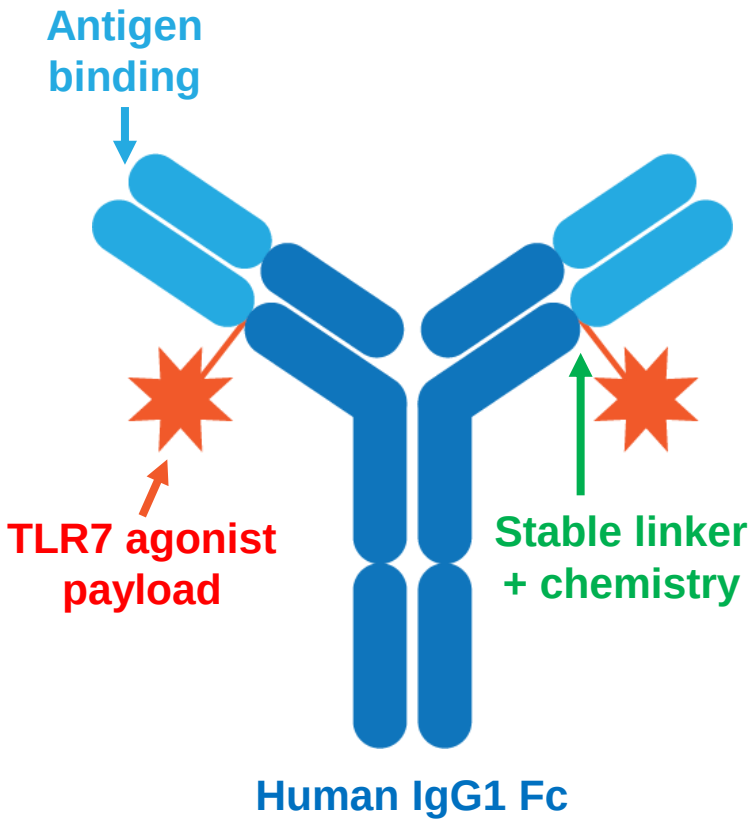
**Toxicity Potential and Reduced Payload Delivery to TME**




## Ambrx ADC and ISAC Focus:

1) Homogenous DAR
2) Stable conjugation chemistry
3) Non-cleavable linker
4) Low payload cell-permeability

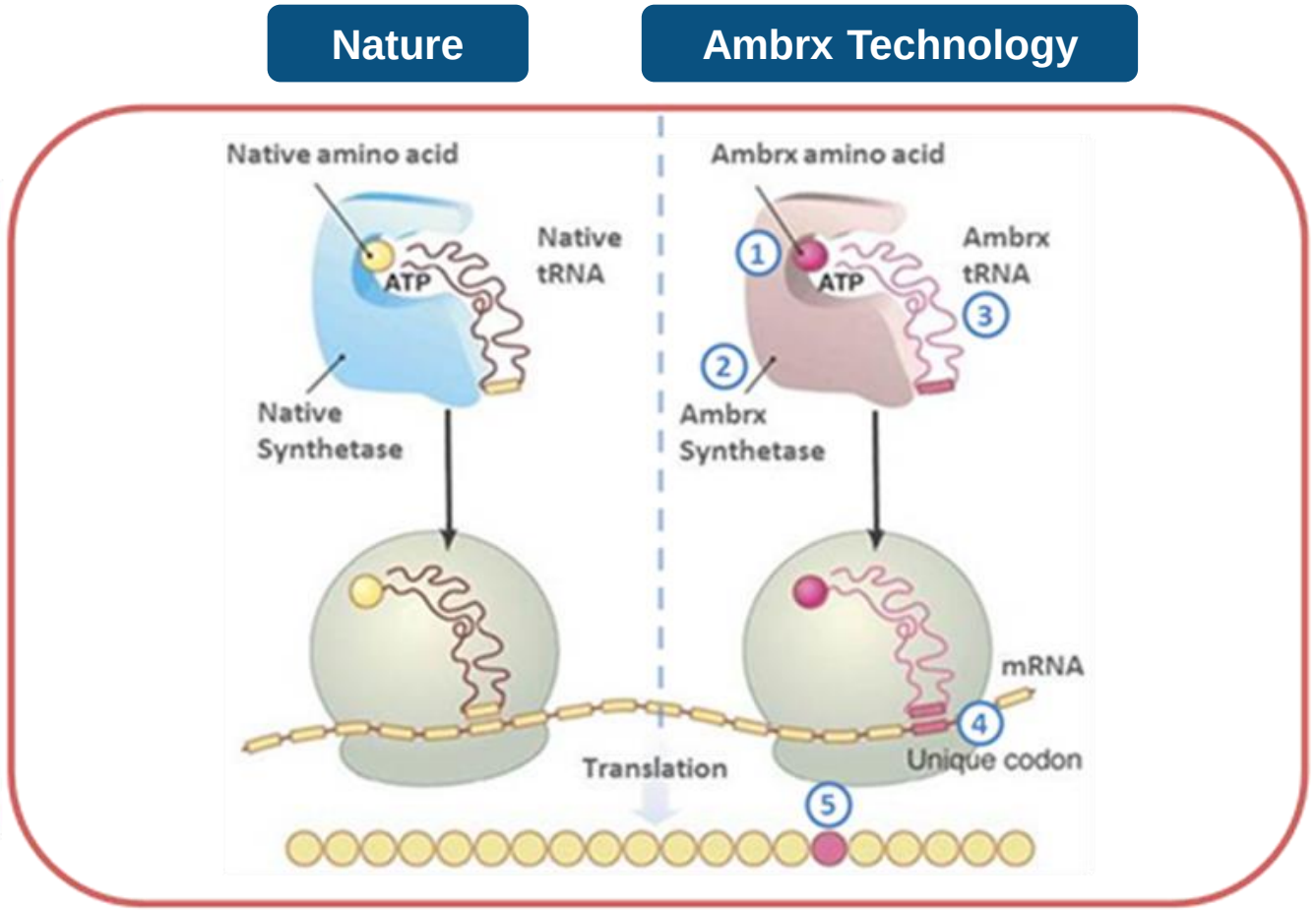
# Ambrx ISAC Platform: Highly Stable and Homogenous via Site-Specific Conjugation and Optimized Linker Chemistry

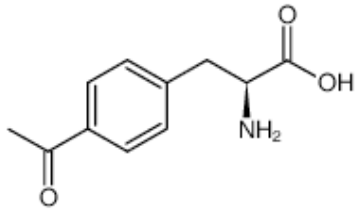



## Multi-Factor Optimization to Design Selective, Potent, and Stable ISACs

- **Antibody:** tumor-associated antigen (TAA) selection, epitope, Fc function
- **Payload**: target, potency, bioconjugation potential, cell permeability
  - **Ambrx: novel TLR7 agonist (TLR7a)** payloads have a range of potencies
  - Free drug-linker has low cell permeability
- **ISAC:** linker stability, DAR, and conjugation site
  - **Site-specific conjugation** enables **high DAR homogeneity**
  - Optimized conjugate site and **stable linker chemistry**
  - Drug-Linker hydrophilicity optimized for enhanced PK

# ReCODE and EuCODE Platforms: An Expanded Genetic Code Incorporating Synthetic Amino Acids (SAAs)

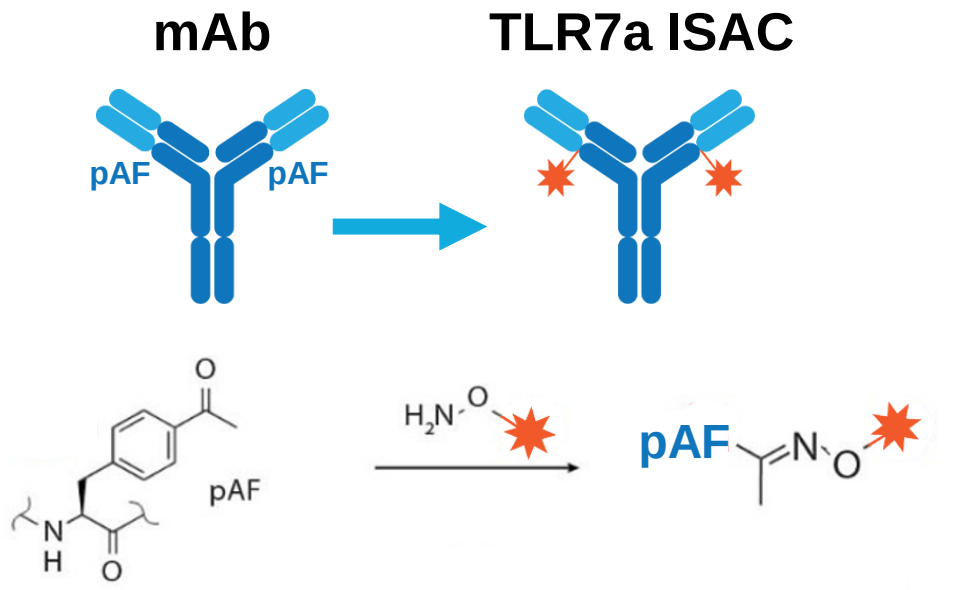


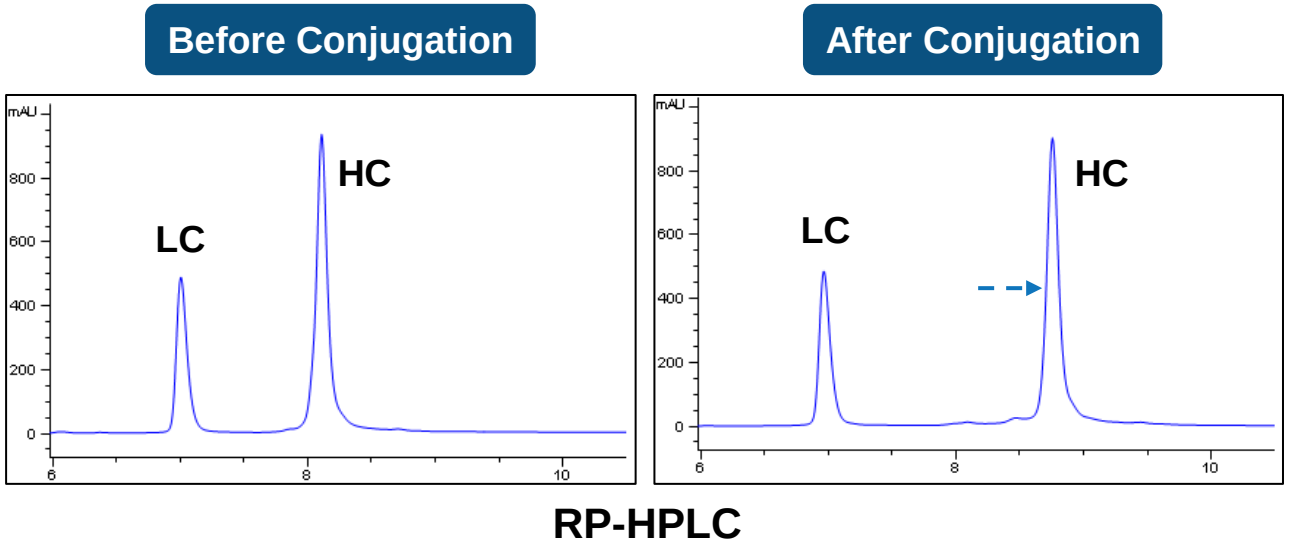




para-Acetyl-phenylalanine

1. Ambrx's SAA: e.g., pAF
2. Orthogonal tRNA synthetase
3. Orthogonal tRNA
4. Unique Amber Codon in mRNA
5. SAA incorporation site on protein

# Ambrx Site-Specific Conjugation Technology Facilitates Highly Homogenous ISAC Generation

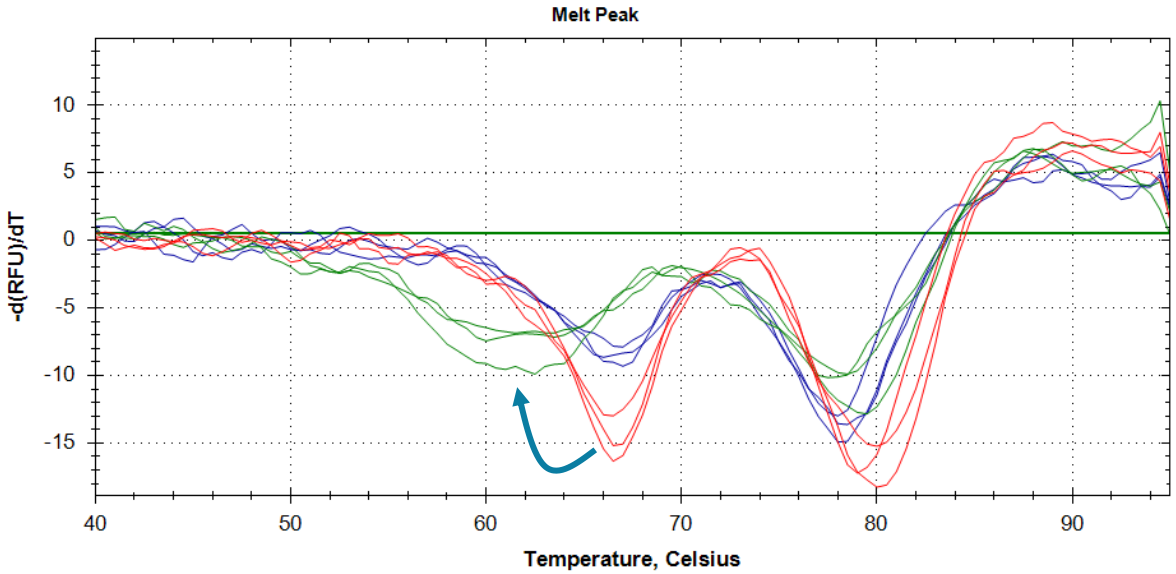



- **Oxime conjugation chemistry**: homogenous, clinically validated stability (ARX788 and ARX517)
- Conjugation site optimized for pharmacologic and biophysical properties
- Focus on **non-cleavable linkers**

# Site-Specific ISAC Conjugates Display Increased Parental mAb-Like Thermal Stability vs. Cysteine Conjugates

**DSF Thermostability Analysis (SYPRO Orange)**

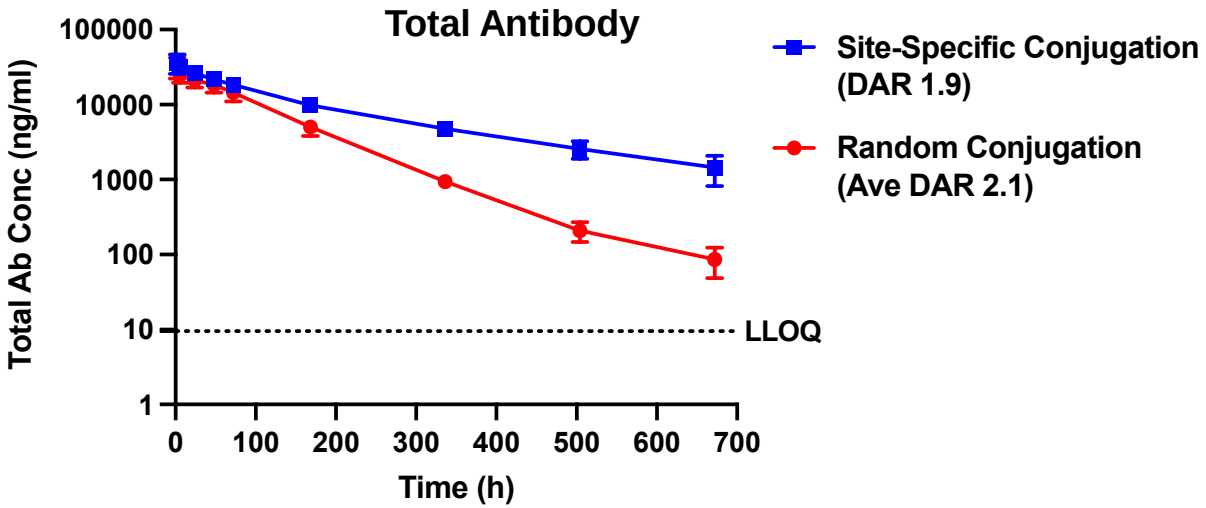



— **Parental mAb**
— **Site-Specific ISAC, Ambrx Drug-Linker (DAR 1.9)**
— **Random Cys ISAC, Ambrx Drug-Linker (DAR 2.1)**
(n=3 measurements)

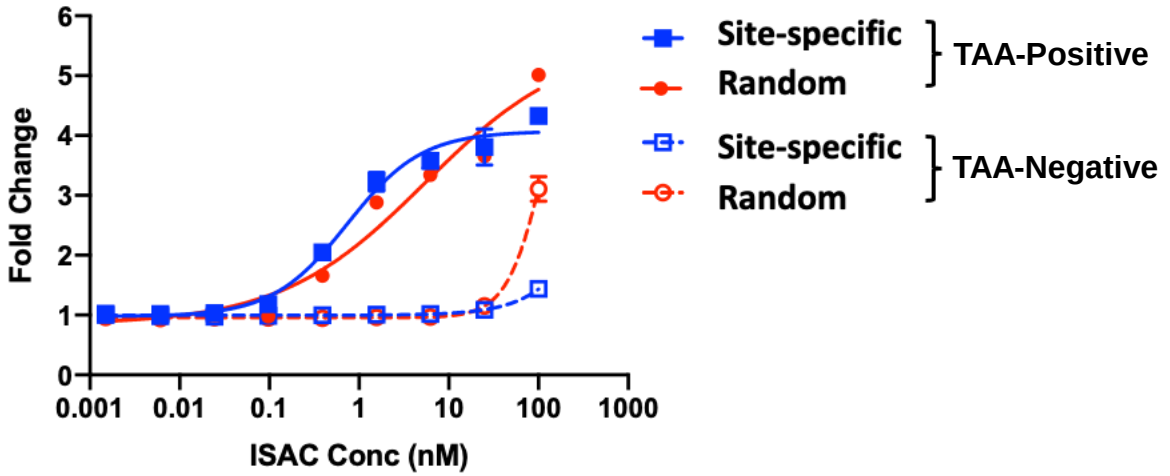
**Ambrx site-specific ISAC has a parental mAb-like CH2 transition temperature, unlike DAR-matched random Cys-conjugate ISAC**

# Same mAb, Same TLR7 Payload: Site-specific Conjugation Promotes Extended PK and Reduces Target-Independent Activity Compared to Cysteine Conjugate

## Pharmacokinetics in Balb/c mice

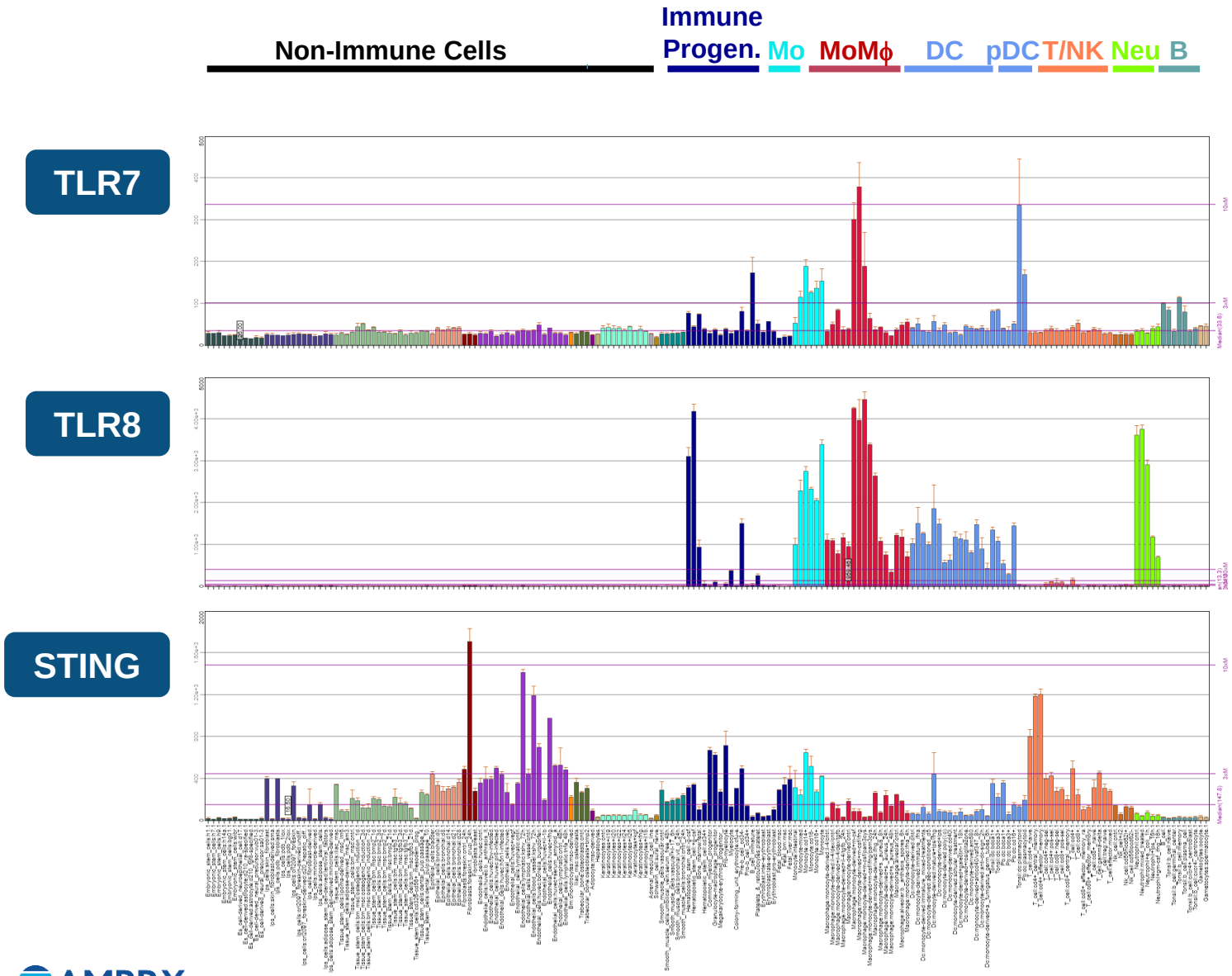



## Tumor cell + macrophage reporter co-culture




**Site-specific conjugation facilitates extended PK and reduced off-target activity vs. Cys-conjugation**

# TLR7 Prioritized as Payload Target Due to Selective Expression and Type I IFN Induction




BioGPS.org, Primary Cell Atlas

## TLR7 Induces Proinflammatory Cytokine and Type I IFN Production by PBMCs

**Inflammatory Mediators**

| Med | TLR7L | TLR8L | TLR9 | Gene |
|---|---|---|---|---|
| 1 | 95 | 297 | 29 | IL6 |
| 1 | 12 | 121 | 15 | IFNG |
| 1 | 11 | 118 | 1 | IL1F9 |
| 1 | 18 | 50 | 2 | IL19 |
| 1 | 20 | 34 | 16 | IL1RN |
| 1 | 8 | 27 | 7 | IL1A |
| 1 | 7 | 23 | 2 | IL10 |
| 1 | 4 | 21 | 3 | TNF |
| 1 | 4 | 10 | 3 | IL2RA |
| 1 | 3 | 9 | 1 | TNFSF9 (41BB-L) |
| 1 | 9 | 8 | 4 | IL15RA |
| 1 | 4 | 8 | 2 | IL27 |
| 1 | 6 | 7 | 2 | TNFAIP6 |
| 1 | 3 | 5 | 1 | TGF-A |
| 1 | 4 | 4 | 3 | IL1B |
| 1 | 2 | 4 | 2 | TNFRSF18 (GITR) |
| 1 | 6 | 3 | 11 | TNFSF10 (TRAIL) |
| 1 | 3 | 0.5 | 3 | TNFSF13B (BAFF) |

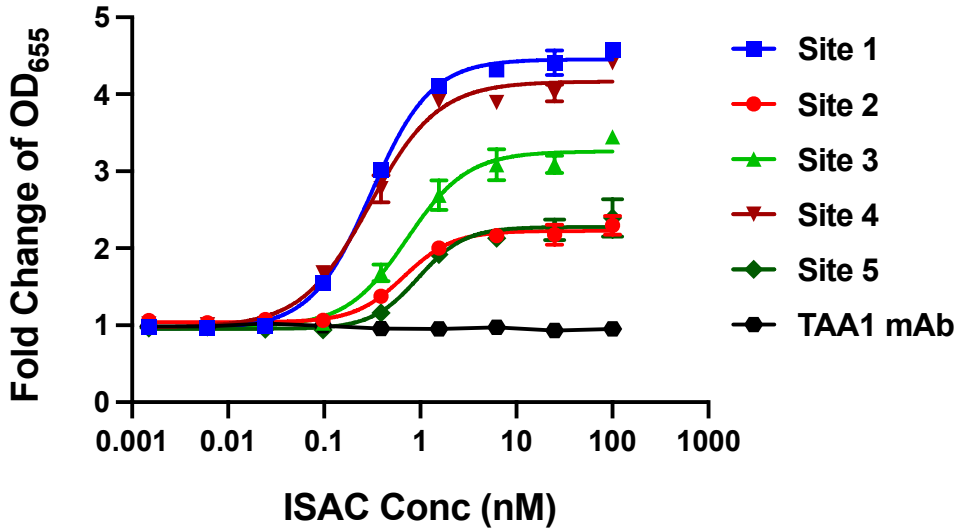
**Type I IFN regulated transcripts**

| Med | TLR7L | TLR8L | TLR9 | Gene |
|---|---|---|---|---|
| 1 | 10 | 3 | 15 | IFIT1 |
| 1 | 10 | 5 | 13 | IFIT3 |
| 1 | 8 | 5 | 9 | ISG15 |
| 1 | 5 | 3 | 8 | IFIT2 |
| 1 | 8 | 3 | 7 | OAS1 |
| 1 | 7 | 4 | 7 | IFI44 |
| 1 | 9 | 4 | 6 | OAS3 |
| 1 | 12 | 5 | 5 | IFI6 |
| 1 | 5 | 3 | 5 | OASL |
| 1 | 5 | 3 | 4 | MX2 |

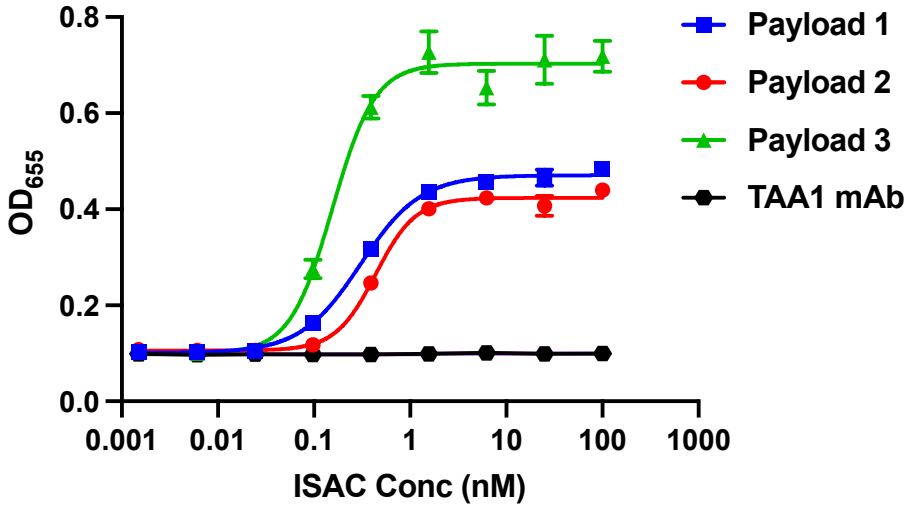
Guiducci et al, *J Exp Med* 2013

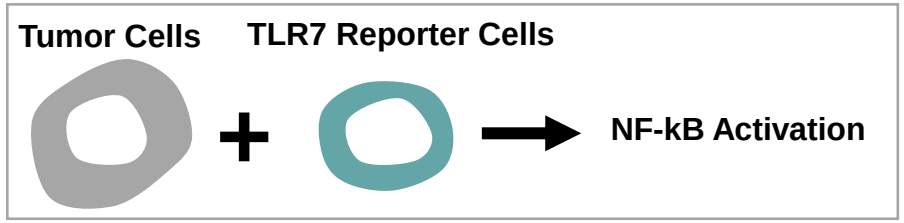
# Different Conjugation Sites and Payloads Yield a Panel of ISACs With Differential Biologic Activity

**Same payload, different conjugation sites**



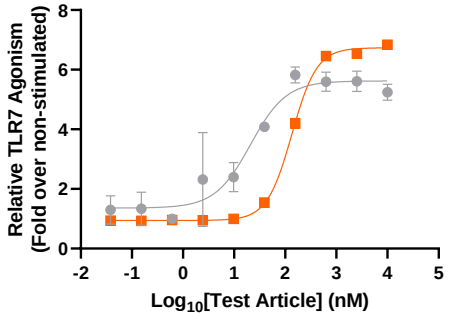

**Same conjugation site, different TLR7a payloads**



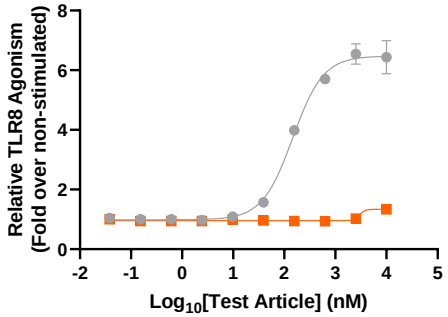



# ARX622- A HER2-Targeted, TLR7-Selective ISAC that Induces Conditional Immune Cell Activation

## Ambrx Drug-Linker is a Novel TLR7-Selective Agonist




TLR7 Reporter Assay

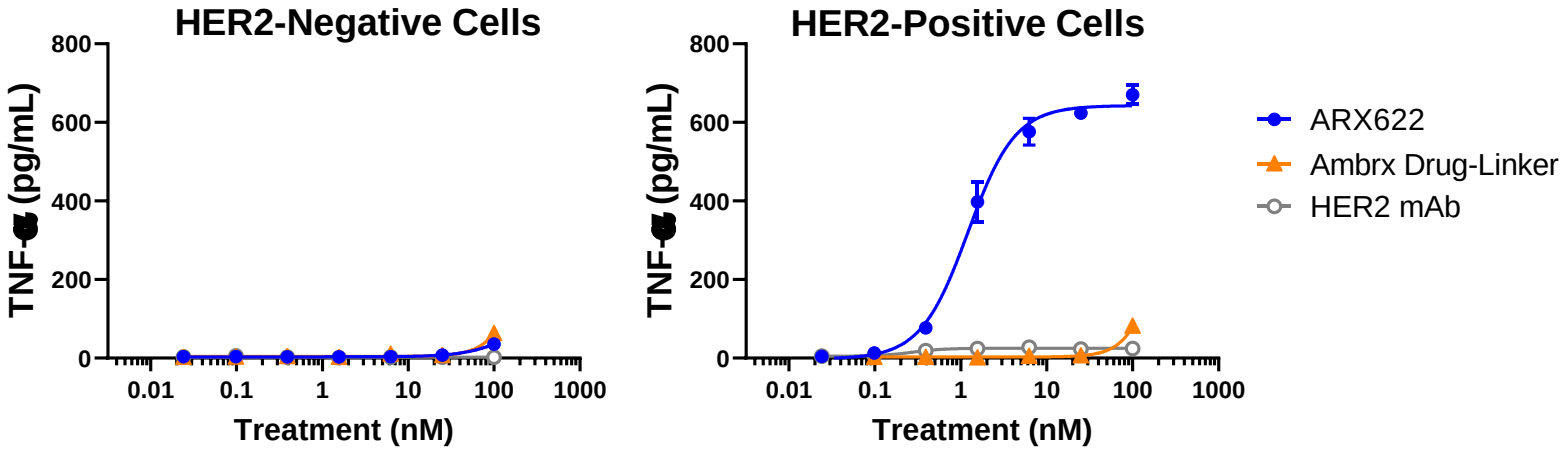


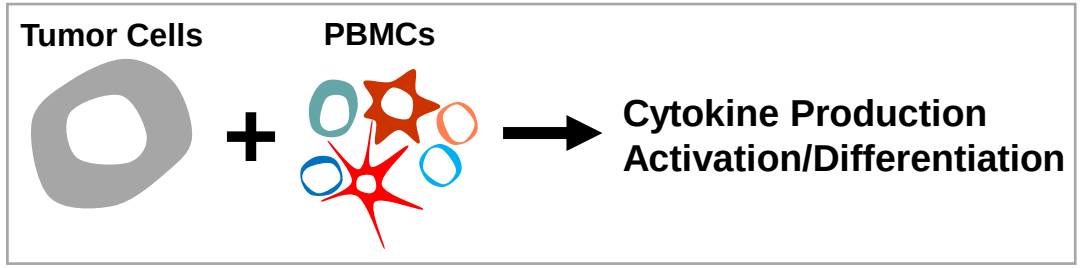

TLR8 Reporter Assay

Positive Control
(TLR7=DSR-6434 / TLR8=R848)

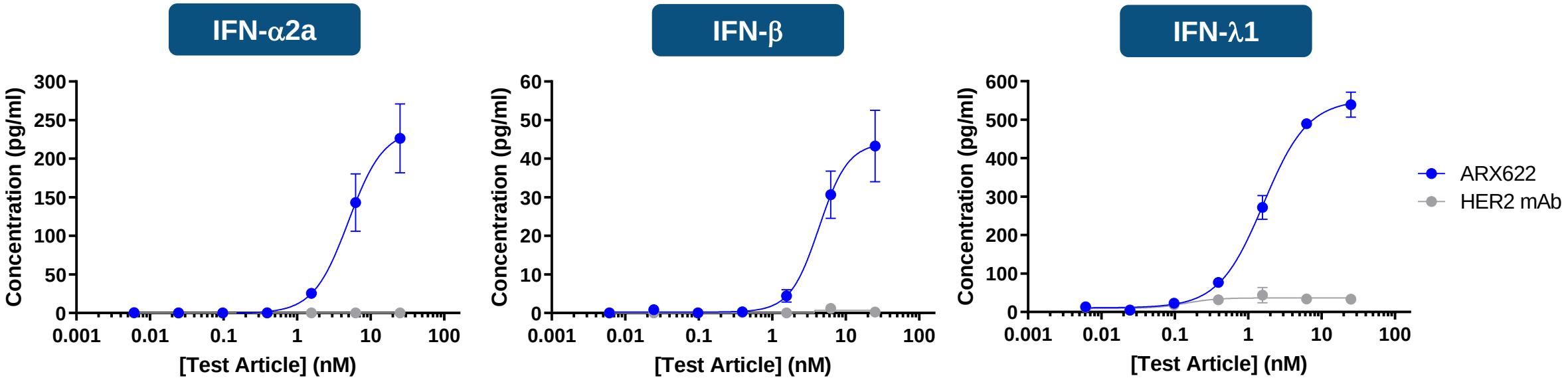
Ambrx ISAC Drug-Linker

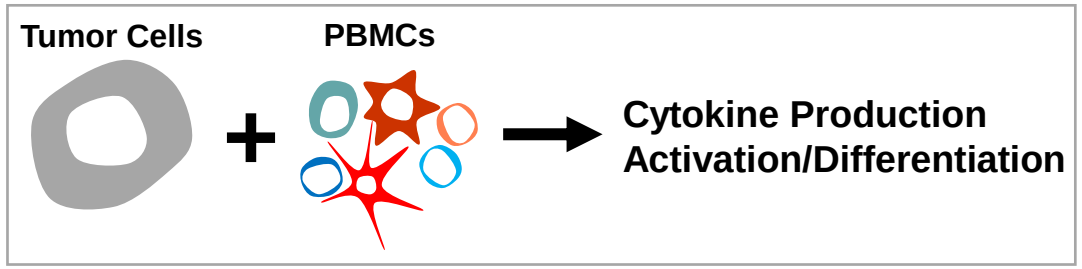
## ARX622 Promotes Conditional Immune Activation in the Presence of HER2+ Tumor Cells



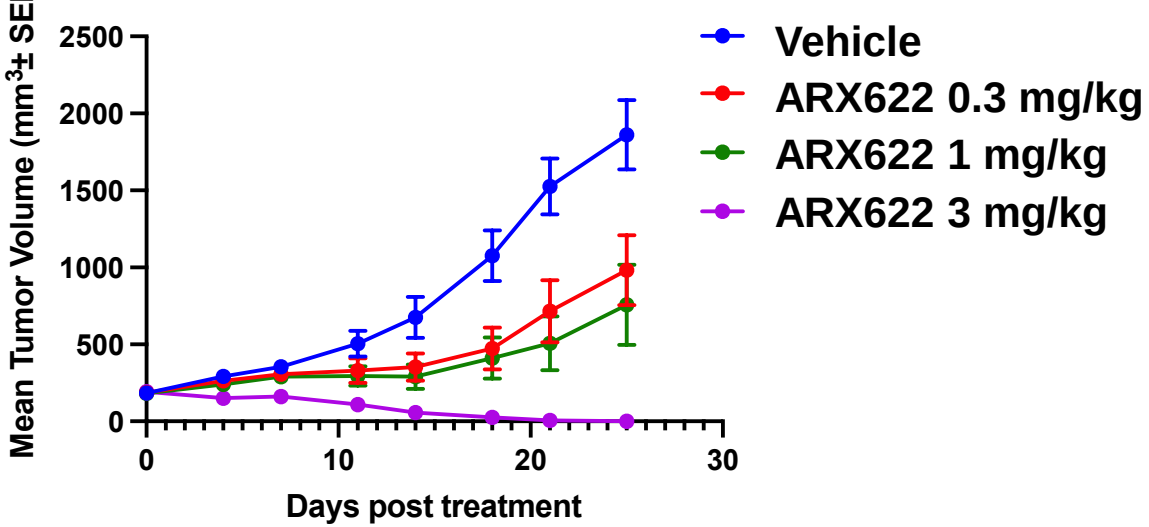
# ARX622 Activates Multiple Anti-Tumor Immune Mechanisms: Robust Type I and Type III IFN Production



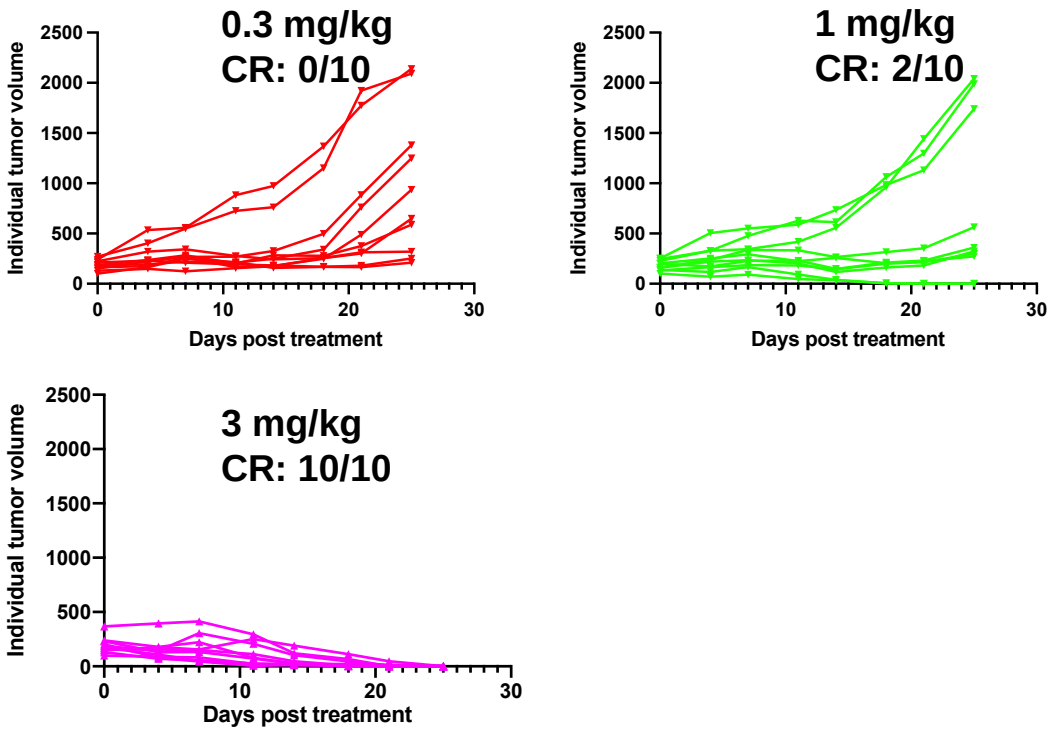



# ARX622 Promotes Dose-Dependent Tumor Growth Inhibition in a Syngeneic Immunocompetent Model

**MC38-hHER2 Cells in Balb/c mice (HER2-low expression)**



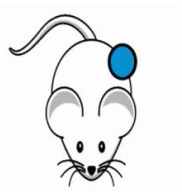

QWx4, i.v.




- ARX622 showed dose-dependent tumor growth inhibition in immunocompetent mice
- No body weight loss or hypersensitivity upon repeated doses

# ARX622 Promotes Tumor-Specific Proinflammatory Cytokine and Type I IFN Production

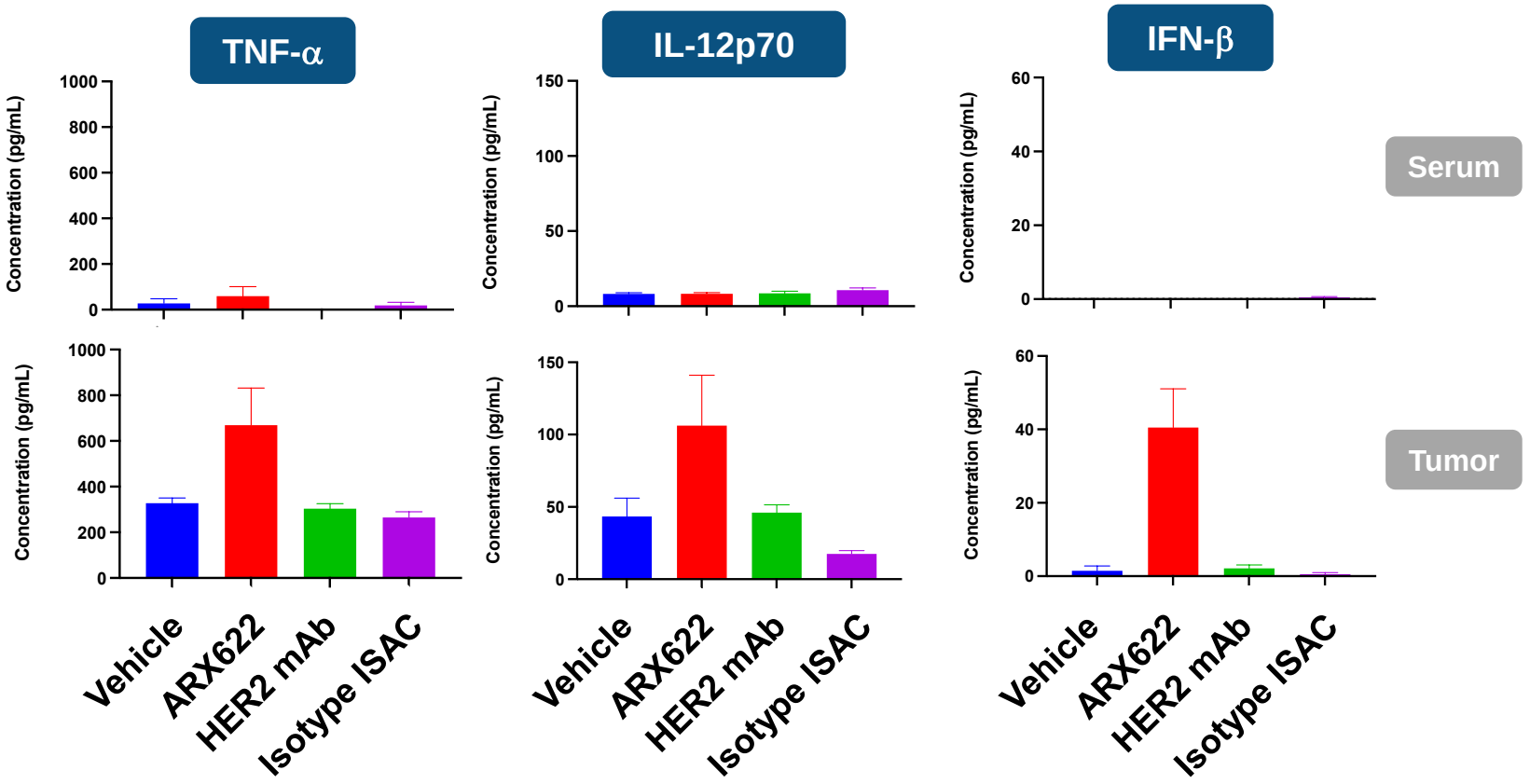
**MC38-hHER2, Balb/c**



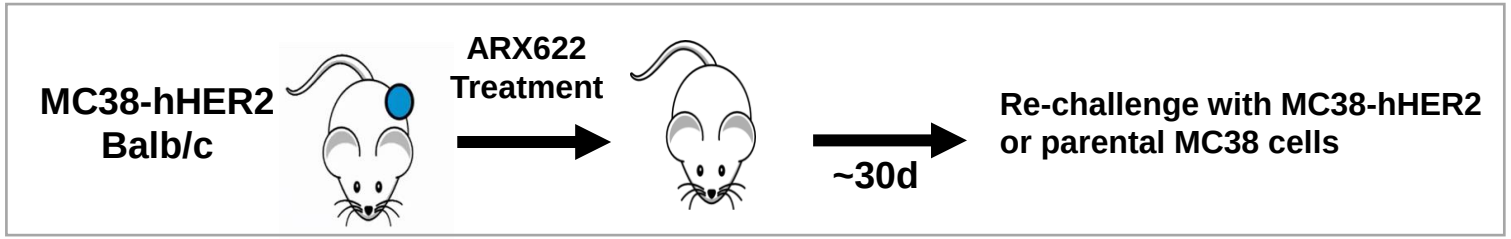
**+ test article**

**24h**

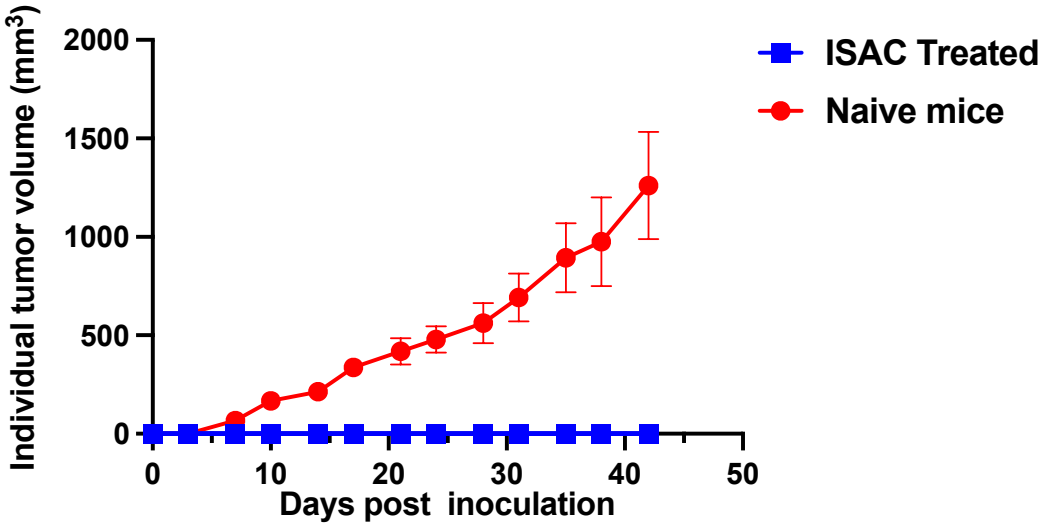
**Tumor cytokines**
**Serum cytokines**



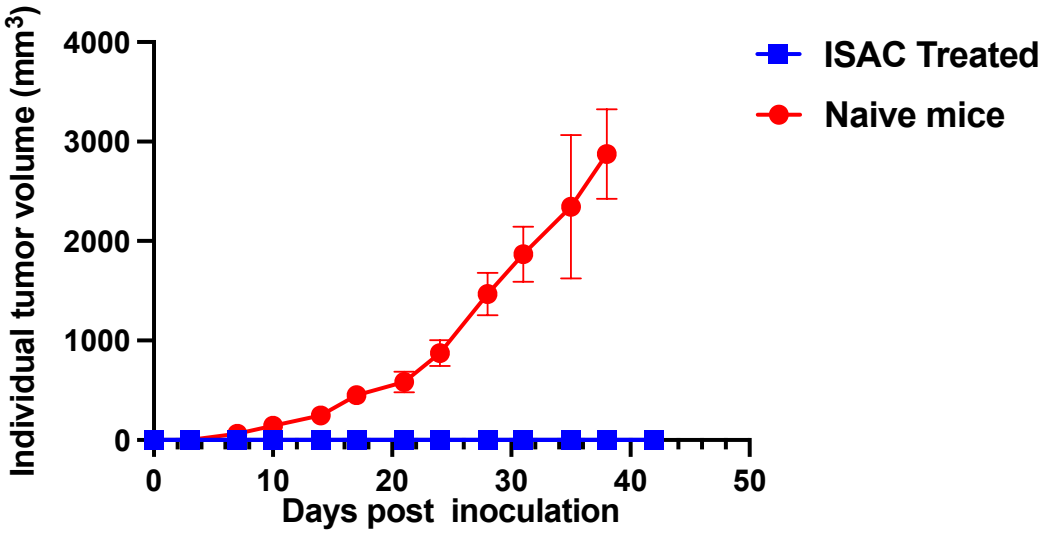
# ARX622 Induces Long Term Protection Against HER2-Negative Tumor Variant Re-Challenge




**Re-challenge: MC38-hHER2 tumor**

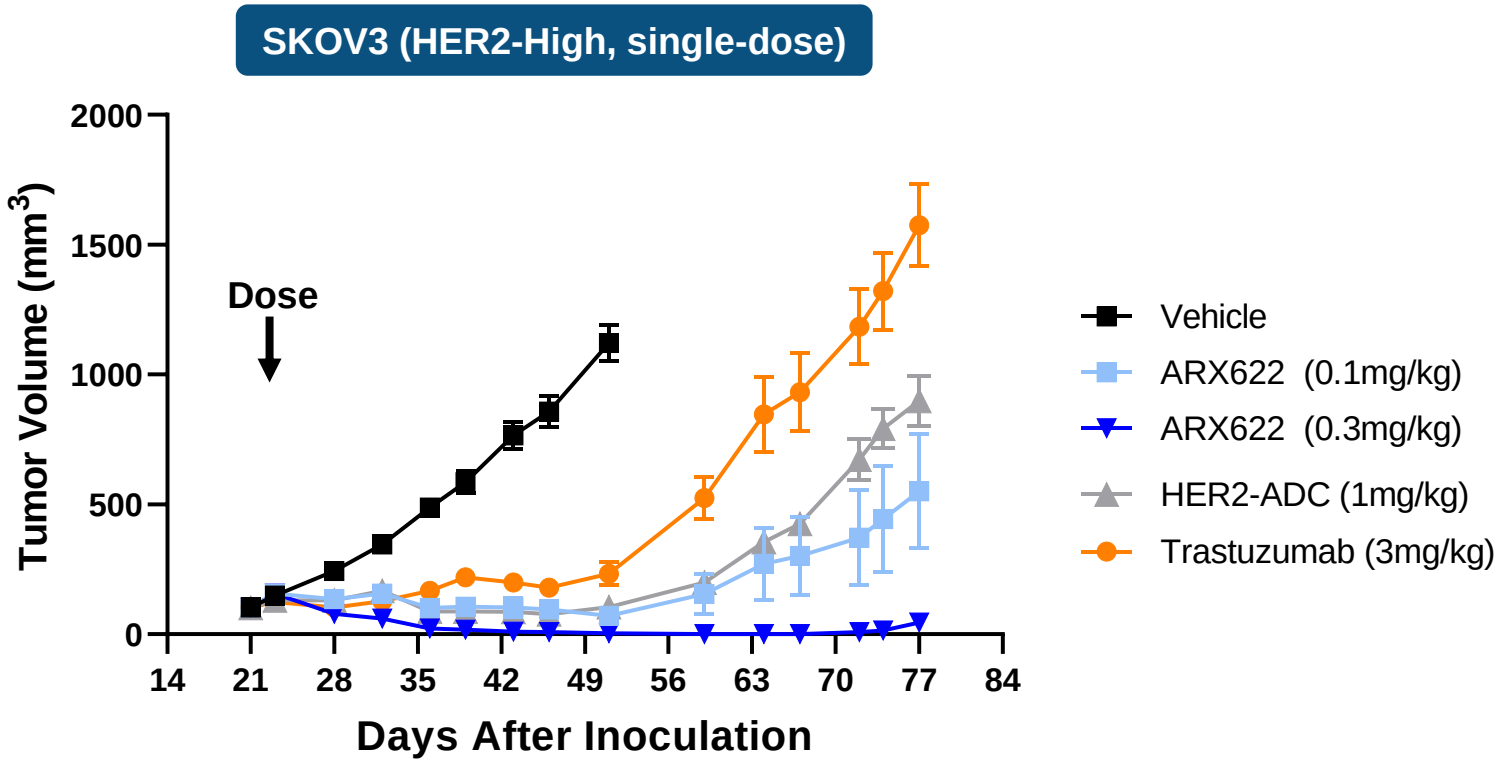



**Re-challenge: MC38 tumor**




**ARX622-treated recipients are protected from HER2-positive and HER2-negative tumor growth**

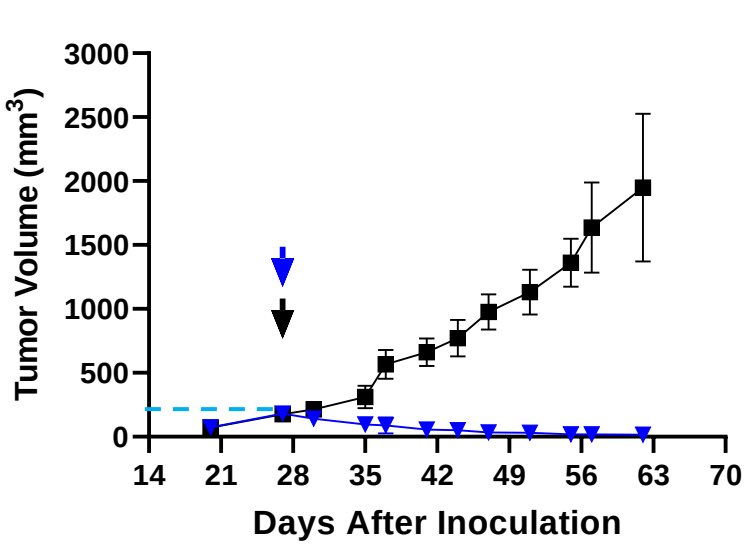
# ARX622 Induces Complete Tumor Regression in the HER2-High SKOV3 CDX Model at Single Doses ≥ 0.3 mg/kg

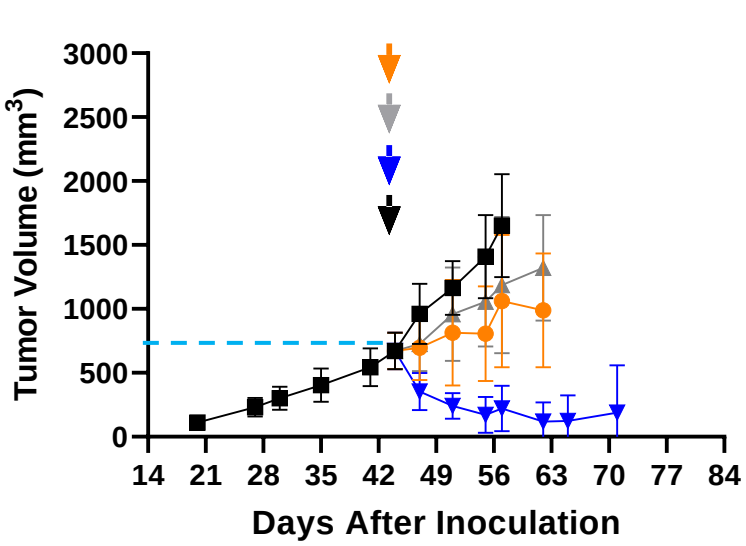


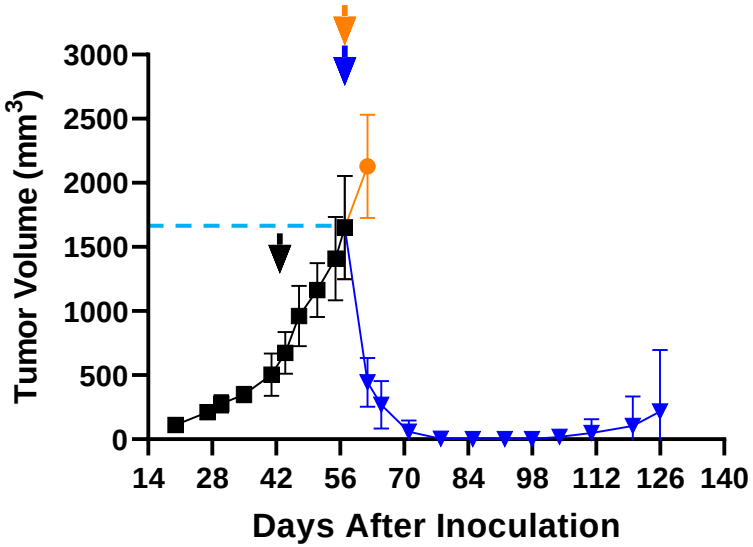

- ISAC exhibited ~10x increased potency vs. MTI-based ADC
- Multiple ISAC mechanisms likely contribute to efficacy: cytostatic cytokines, ADCP, pDC cytotoxicity

# Raising the Bar: Ambrx HER2 ISAC Promotes Regression of Large, Established SKOV3-*scid* Xenograft Tumors

**Dose at:** 200mm³ | 750mm³ | 1700mm³



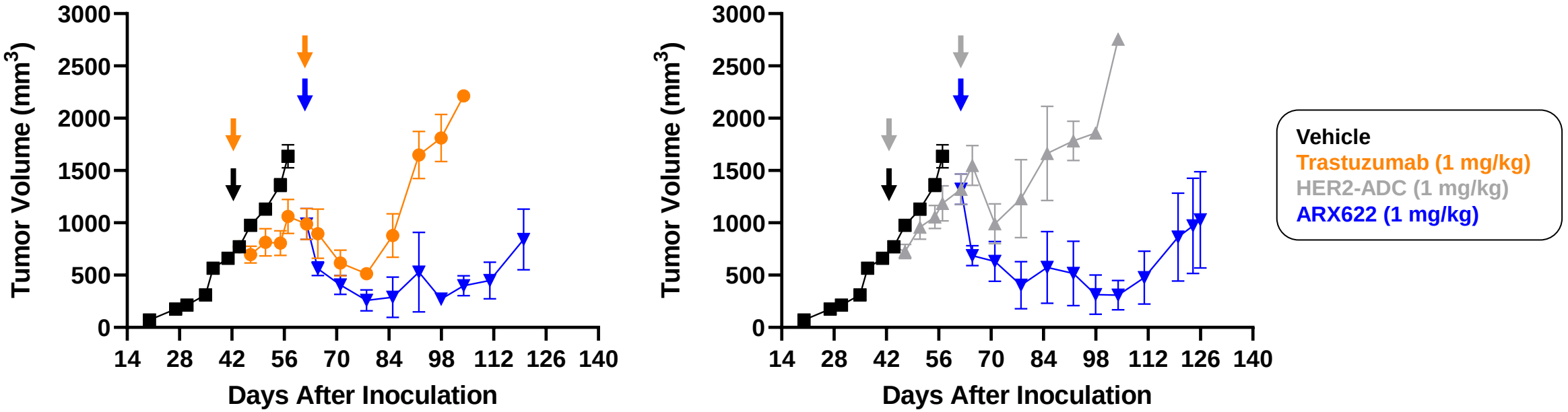




Vehicle
ARX622 (0.3 mg/kg)

Vehicle
Trastuzumab (1 mg/kg)
HER2-ADC (1 mg/kg)
ARX622 (1 mg/kg)

**ARX622 displays anti-tumor activity in therapeutic setting**

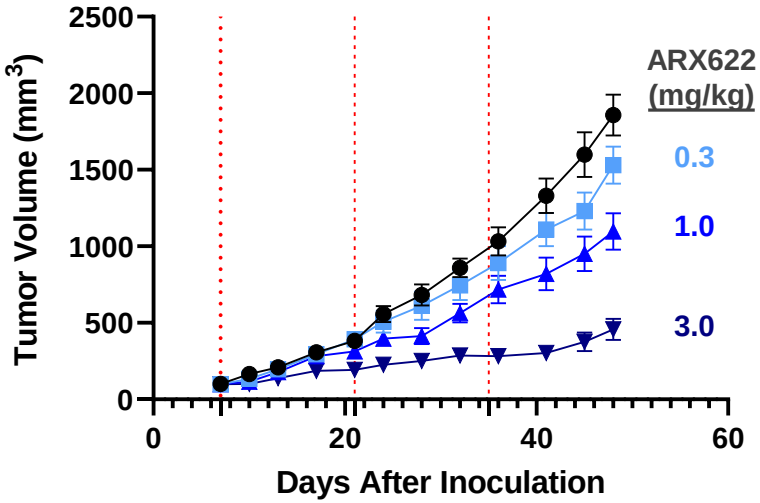
# Ambrx ISAC Reduces SKOV3 Tumor Growth in HER2 mAb and HER2-ADC Non-Responder Mice



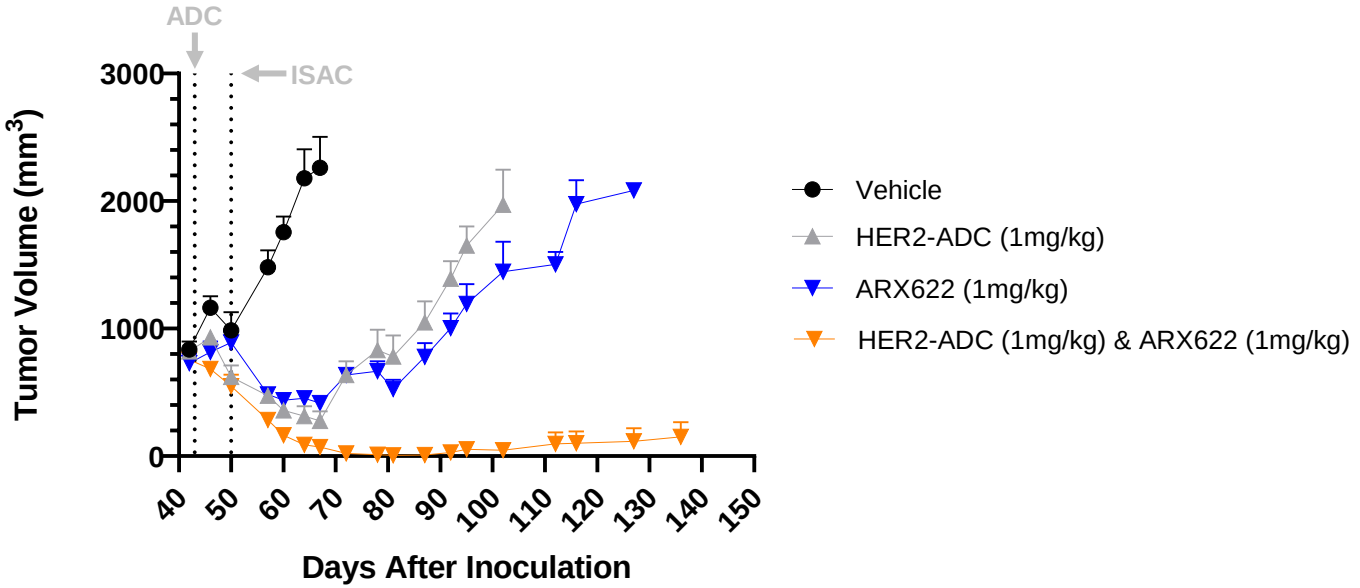

**ARX622 displays anti-tumor activity in HER2-targeted therapy non-responder mice**

# ARX622 + HER2-ADC Combination Induces Enhanced Tumor Growth Inhibition in the HER2-Low JIMT-1 CDX Model

**JIMT-1 (HER2-Low)**




Q2Wx3, i.v.

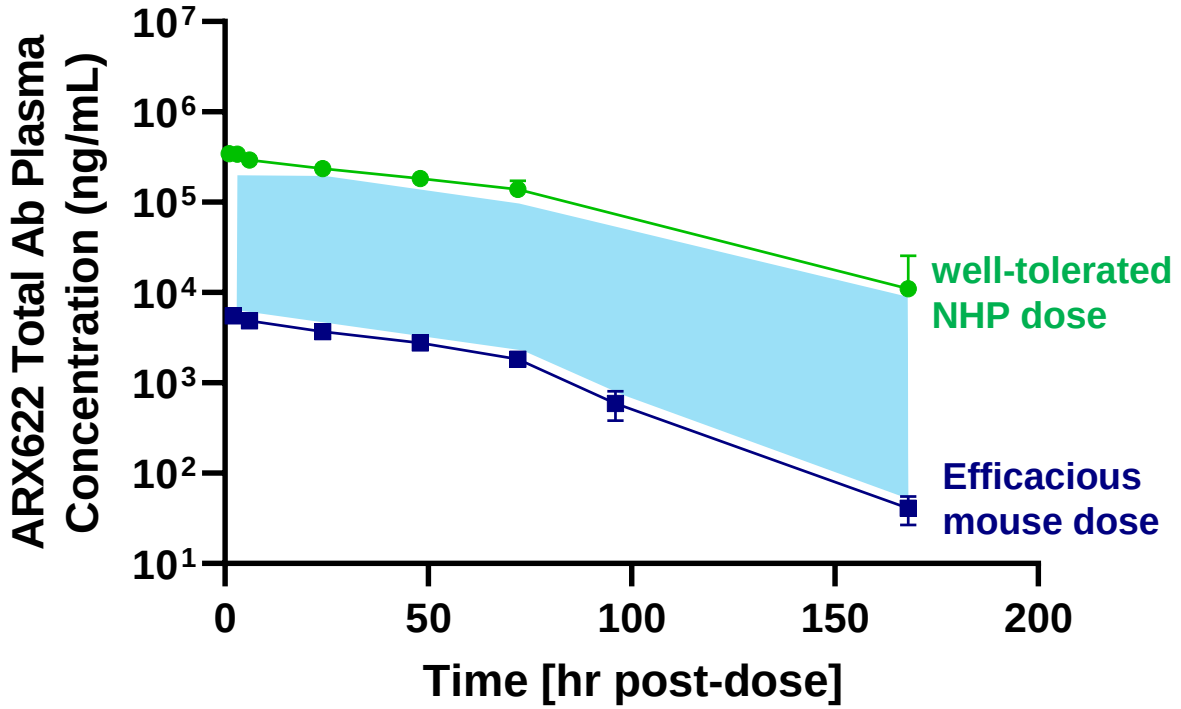



**CDX Summary: Pre-clinical support for monotherapy, combo with HER2-ADCs, and usage post-HER2-ADCs**

# ARX622 is Well-Tolerated in NHPs and Displays ~60x Pre-Clinical Therapeutic Index

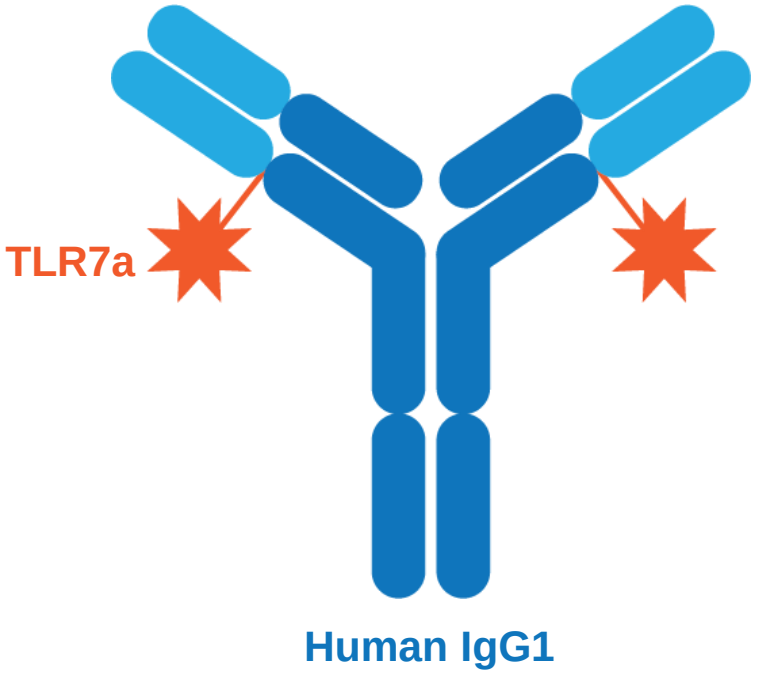
**NHP Study Summary:**
**(repeat i.v. dosing)**

- No mortality or moribundity
- No clinical signs
- No adverse histopathologic findings
- No adverse clinical chemistry or hematologic findings
- Pharmacodynamic Biomarkers: transient cytokine elevation



# Ambrx ISAC Platform is Engineered for Stability and Broad Danger Signal Induction, Facilitating an Encouraging Therapeutic Index and Robust Efficacy Profile

**ARX622: anti-HER2 ISAC**




**Pre-Clinical ARX622 Data Highlights**

- Induction of **broad danger signals**: proinflammatory cytokines, Type I/III IFN, (APC maturation, ADCC enhancement not shown today)
- Repeat doses were **well-tolerated in NHP**
- Robust tumor growth inhibition in syngeneic tumor model with evidence of polyclonal **immunologic memory**
- Complete regression of **large tumors** with single-dose (including post-HER2-ADC treatment)
- Monotherapy **efficacy in HER2-low** setting
- Support for combination with HER2-ADCs