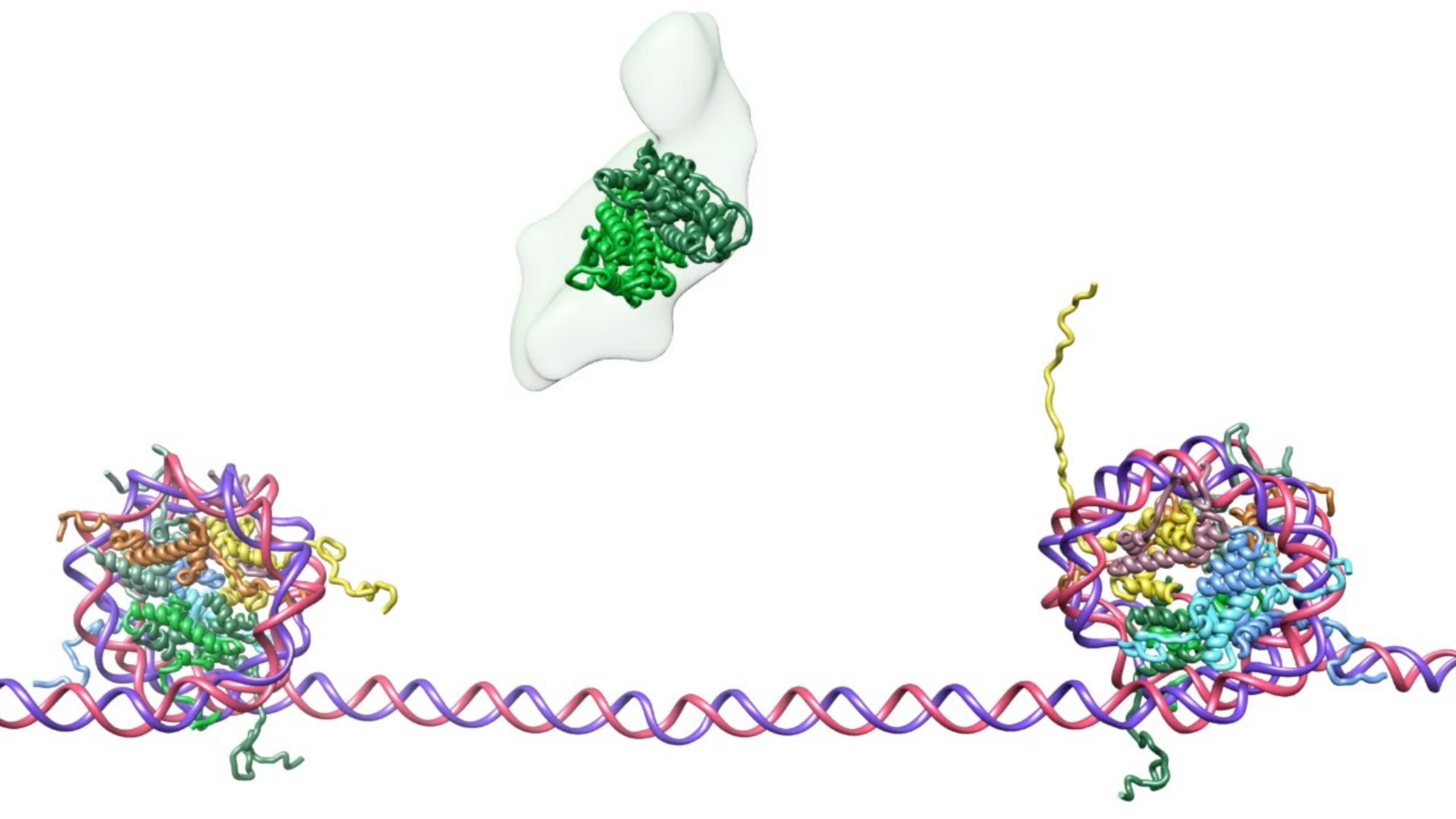
STANFORD
CANCER INSTITUTE

# Drug Development and Future Targeted Agents for Breast Cancer

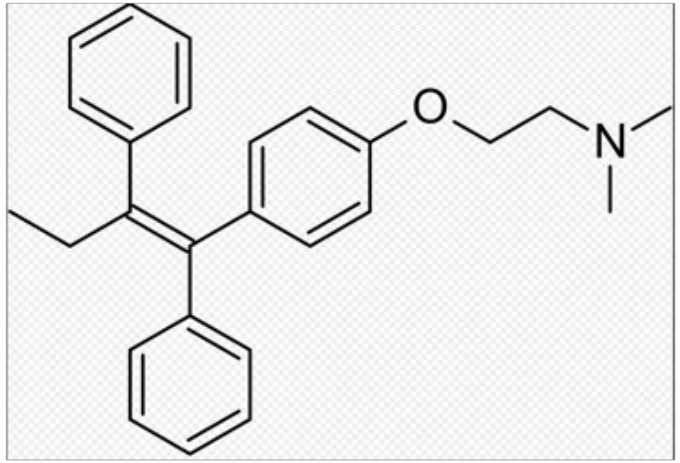
July 2023

Mark Pegram, M.D.
Susy Yuan-Huey Hung Professor of Oncology
Medical Director, Clinical and Translational Research Unit
Associate Dean for Clinical Research Quality
Stanford University School of Medicine



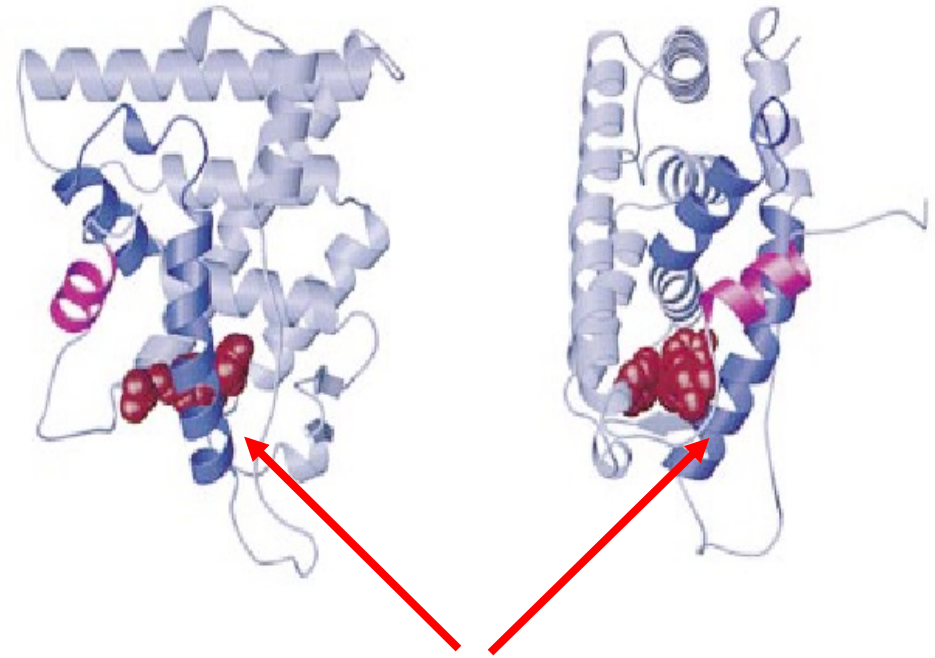
# SERM Tamoxifen binding to ER alters conformation of helix 12 and disrupts interaction with ER co-activators

ICI-46,474




-- First synthesized 1962 as a failed contraceptive
-- active metabolites = 4-OHTAM and endoxifen
-- FDA approval 1977
-- > 2 million prescriptions/yr

ERα/Tamoxifen

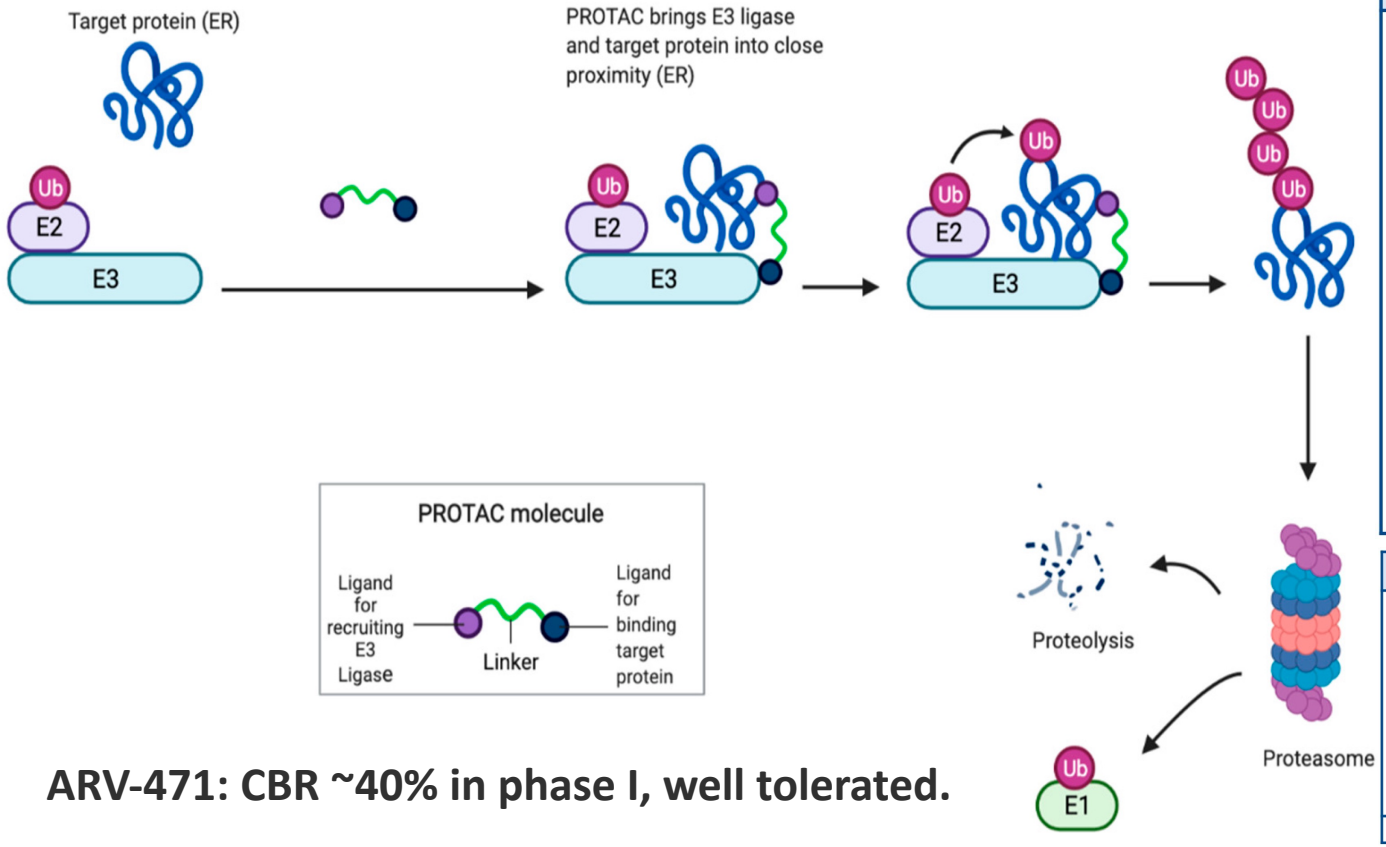


Helix 12

# PROTACs

PROTACs: proteolysis targeting chimeras are heterobifunctional molecules made up of a ligand for ER (target protein) and another ligand, serving as the E3 ubiquitin ligase complex substrate.

Once PROTACs bind to ER, recruit the E3 ubiquitin ligase complex, leading to a polyubiquitilation of ER ending on a proteasomal degradation.




**ARV-471: CBR ~40% in phase I, well tolerated.**

**Figure 2: Inhibition of breast cancer cell growth with vepdegestrant plus abemaciclib (A) or ribociclib (B) vs fulvestrant in preclinical models[9]**

Layman RH, et al. Journal of Clinical Oncology 2023 41:16_suppl, TPS1121-TPS1121.

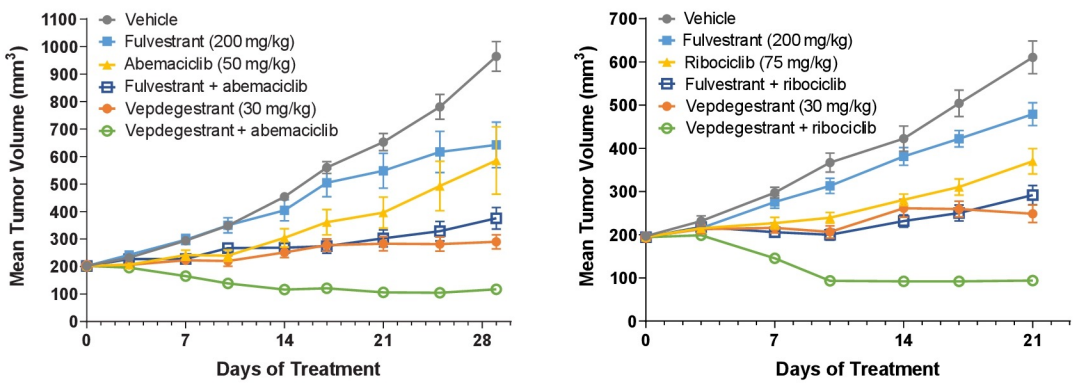
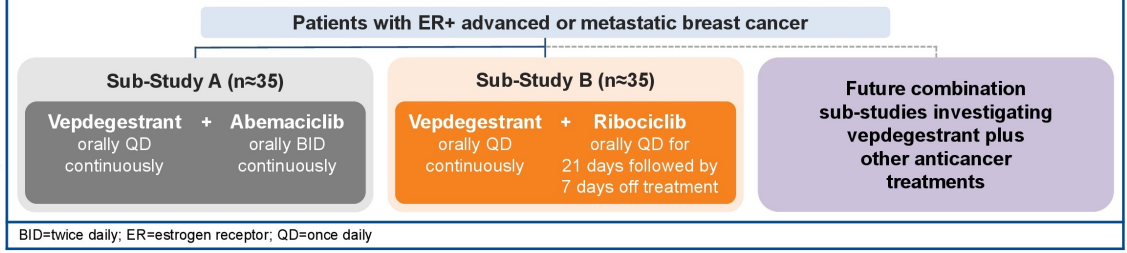



**Figure 3: TACTIVE-U trial schema**





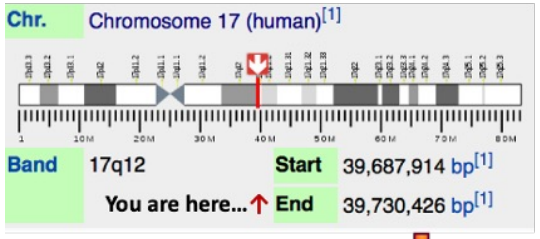

Hamilton E, et al. Journal of Clinical Oncology 2022 40:16_suppl, TPS1120-TPS1120.

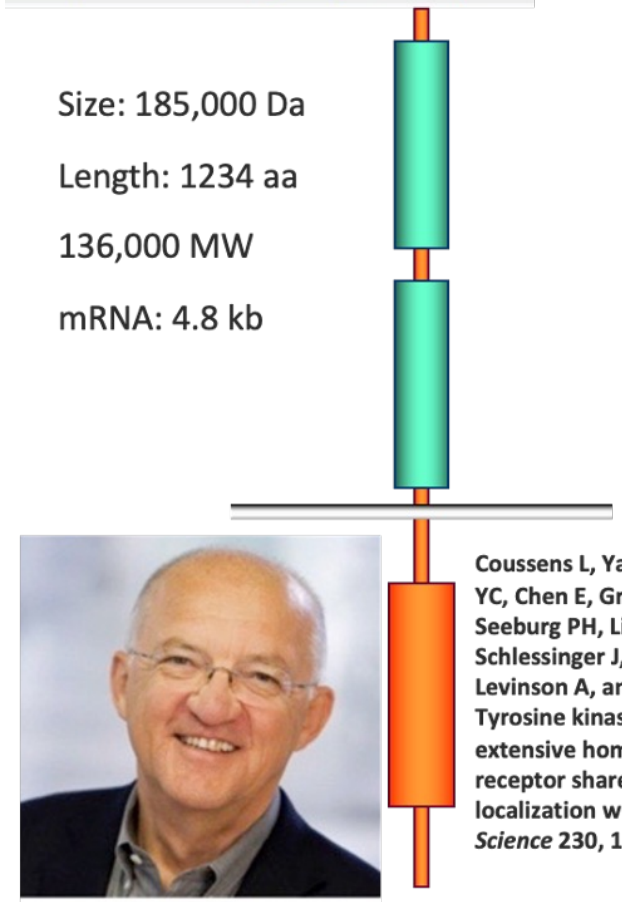
Hernando C et al, IJMS 2021.

# 2023 = 25th Anniversary of Trastuzumab

## 2019 Lasker DeBakey Clinical Medical Research Award

**"For invention of a targeted antibody therapy for breast cancer"**

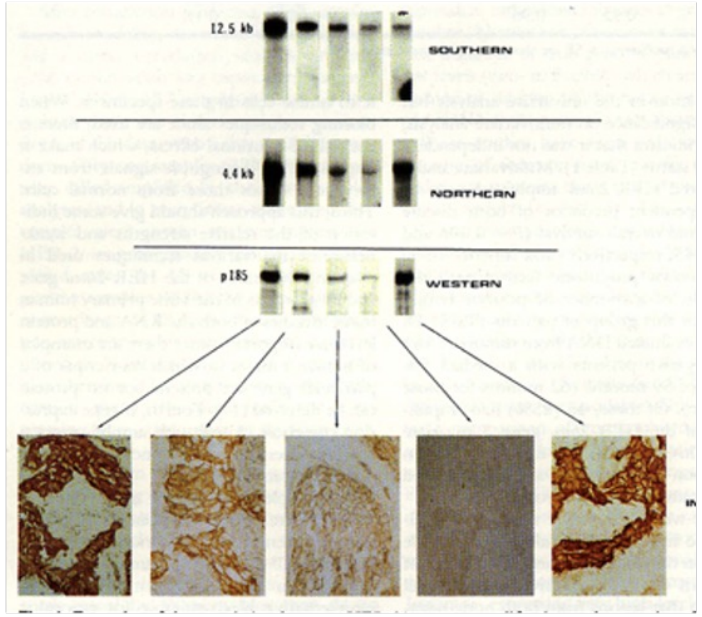




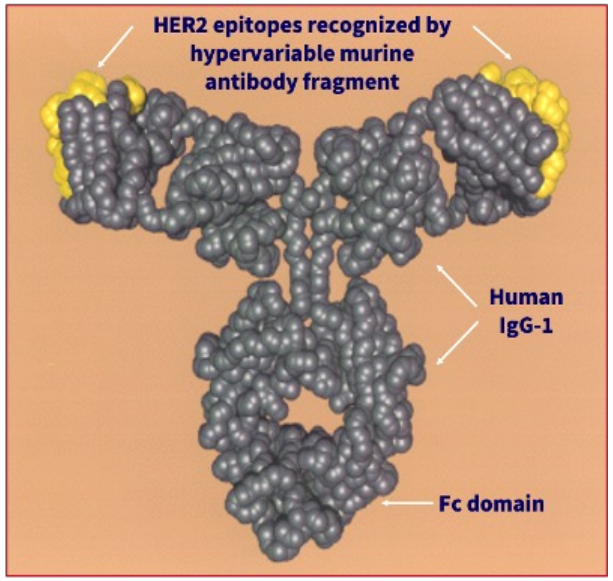

Axel Ullrich
Max Planck Institute of Biochemistry

Coussens L, Yang-Feng TL, Liao YC, Chen E, Gray A, McGrath J, Seeburg PH, Libermann TA, Schlessinger J, Francke U, Levinson A, and Ullrich A. (1985) Tyrosine kinase receptor with extensive homology to EGF receptor shares chromosomal localization with neu oncogene. *Science* 230, 1132-1139




Dennis J. Slamon
University of California, Los Angeles

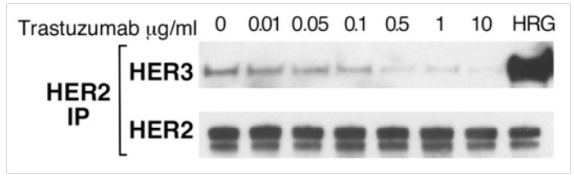
Slamon *et al*. Science 1987 & 1989

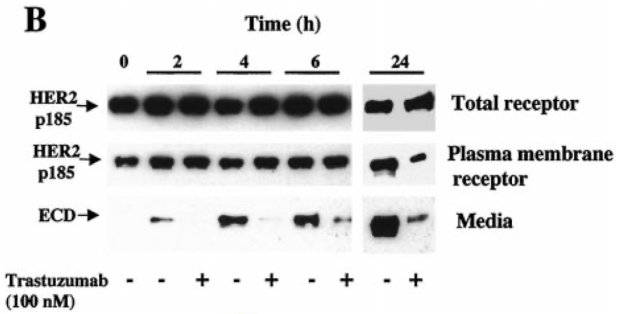


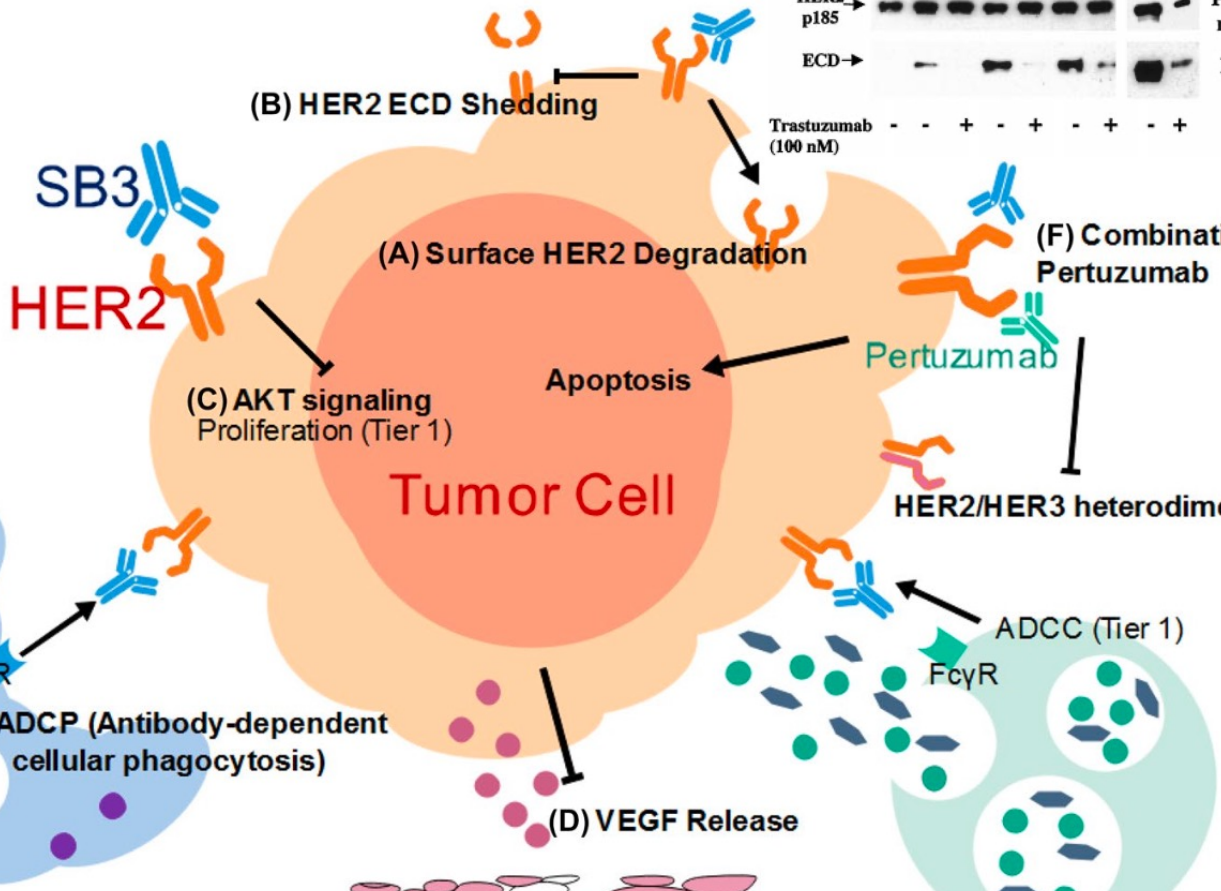

H. Michael Shepard
Genentech

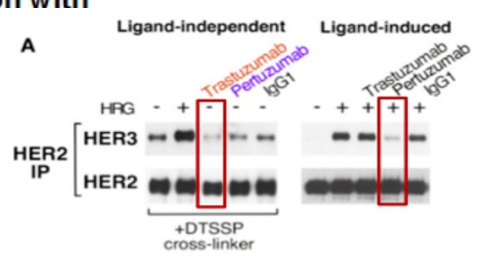
Carter P, et al. Proc Natl Acad Sci USA 89: 4285-89 (1992).

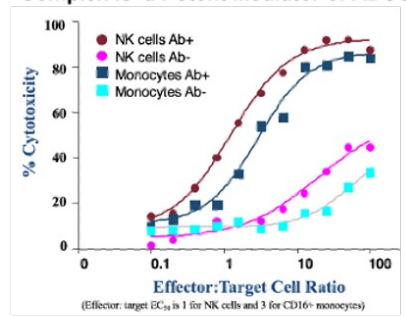
# Overview of the mechanism of action of trastuzumab on in vitro mono- and combination therapy



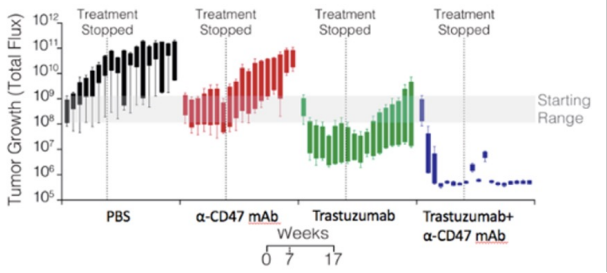



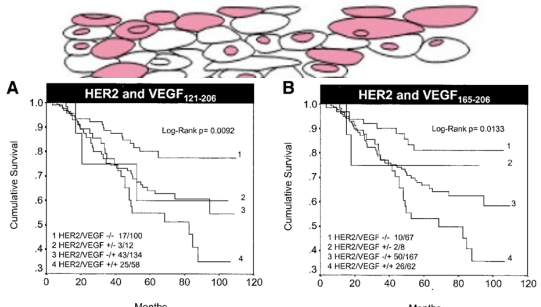



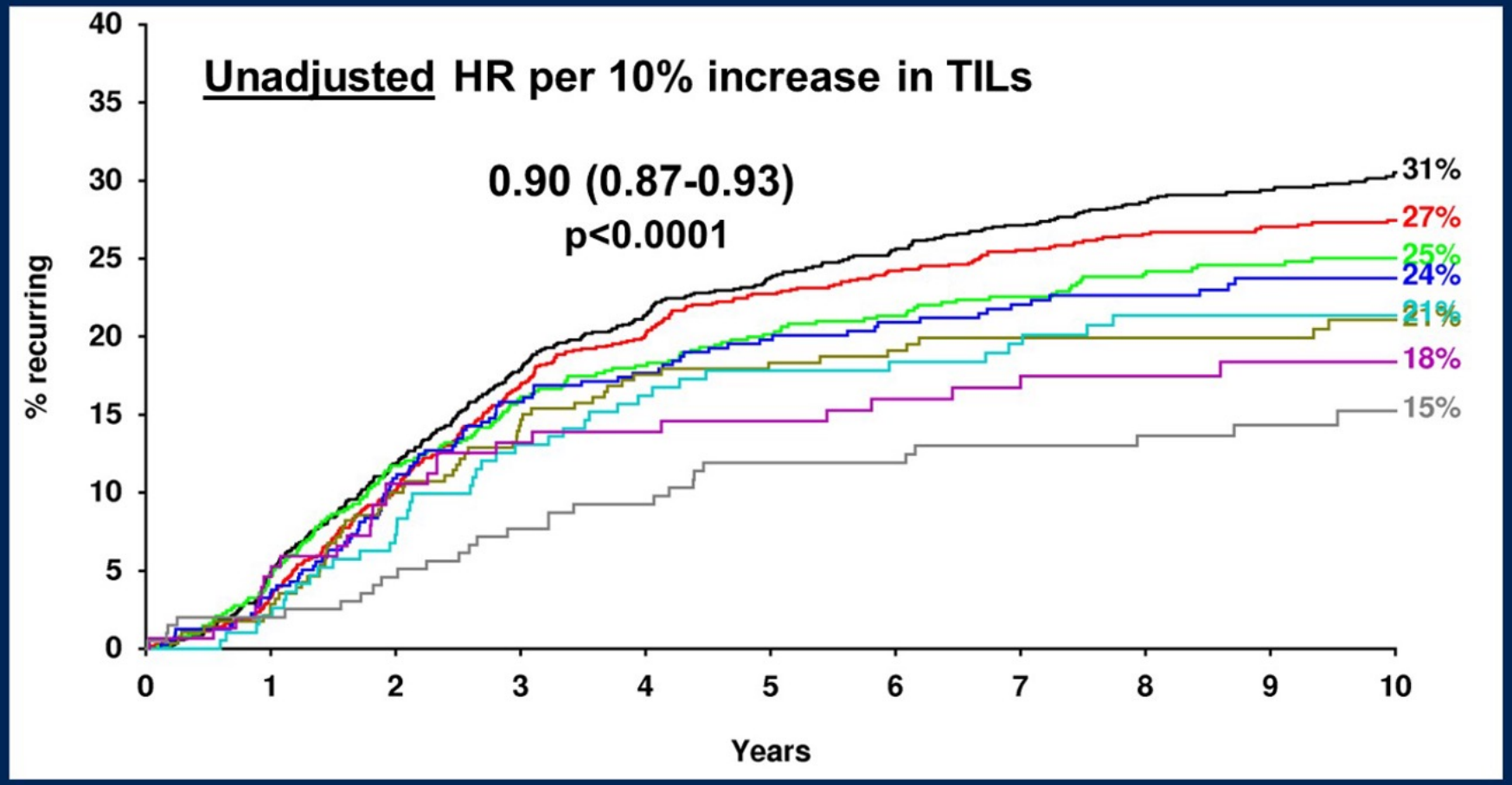

Pegram, et al., Proc Am Assoc Cancer Res 38: 602, 1997 (abstr 4044).

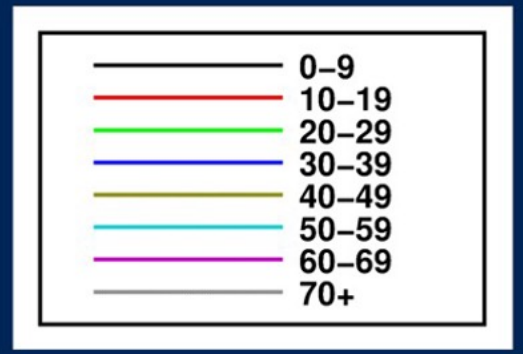



Kyungyeol Paek, et al. BioDrugs (2019) 33:661–671.

# Investigating prognostic effect of TILs on recurrence



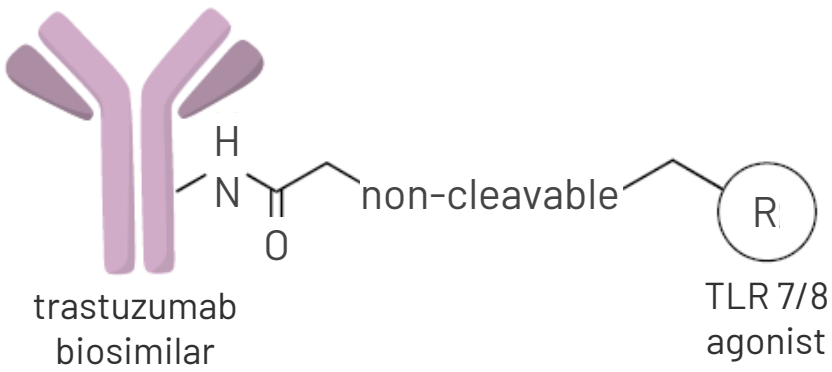




| | No. Patients | No. Events |
|---|---|---|
| 0–9 | 1252 | 372 |
| 10–19 | 1068 | 293 |
| 20–29 | 617 | 153 |
| 30–39 | 401 | 90 |
| 40–49 | 282 | 58 |
| 50–59 | 193 | 40 |
| 60–69 | 154 | 28 |
| 70+ | 201 | 28 |

# Novel, First-in-Class Boltbody™ Immune-Stimulating Antibody Conjugate (ISAC)

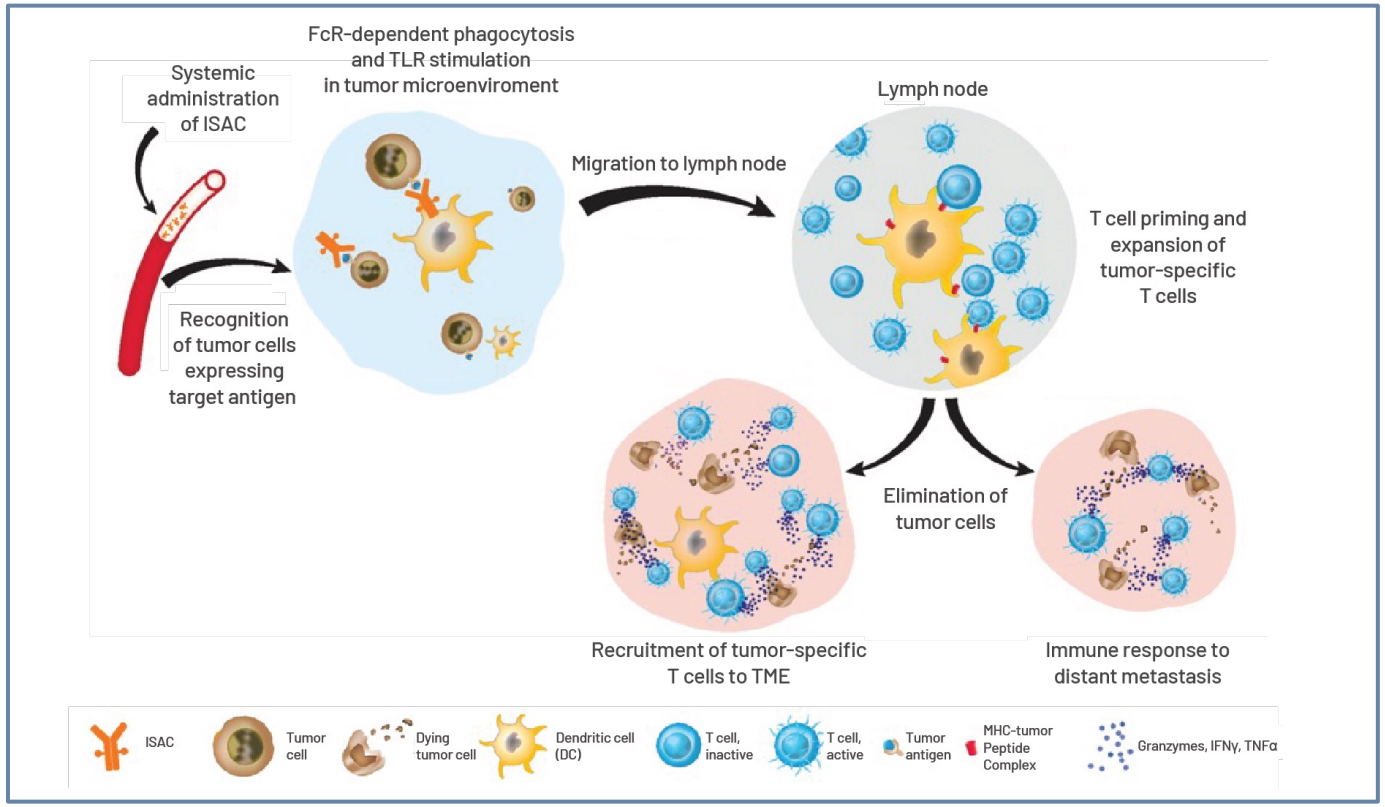
## Molecular Structure

- BDC-1001 consists of
  - Trastuzumab biosimilar
  - Payload: TLR7/8 agonist
  - Linker: non-cleavable
- BDC-1001 linker-payload is cell membrane-impermeable




## Proposed Mechanism of Action (MOA)

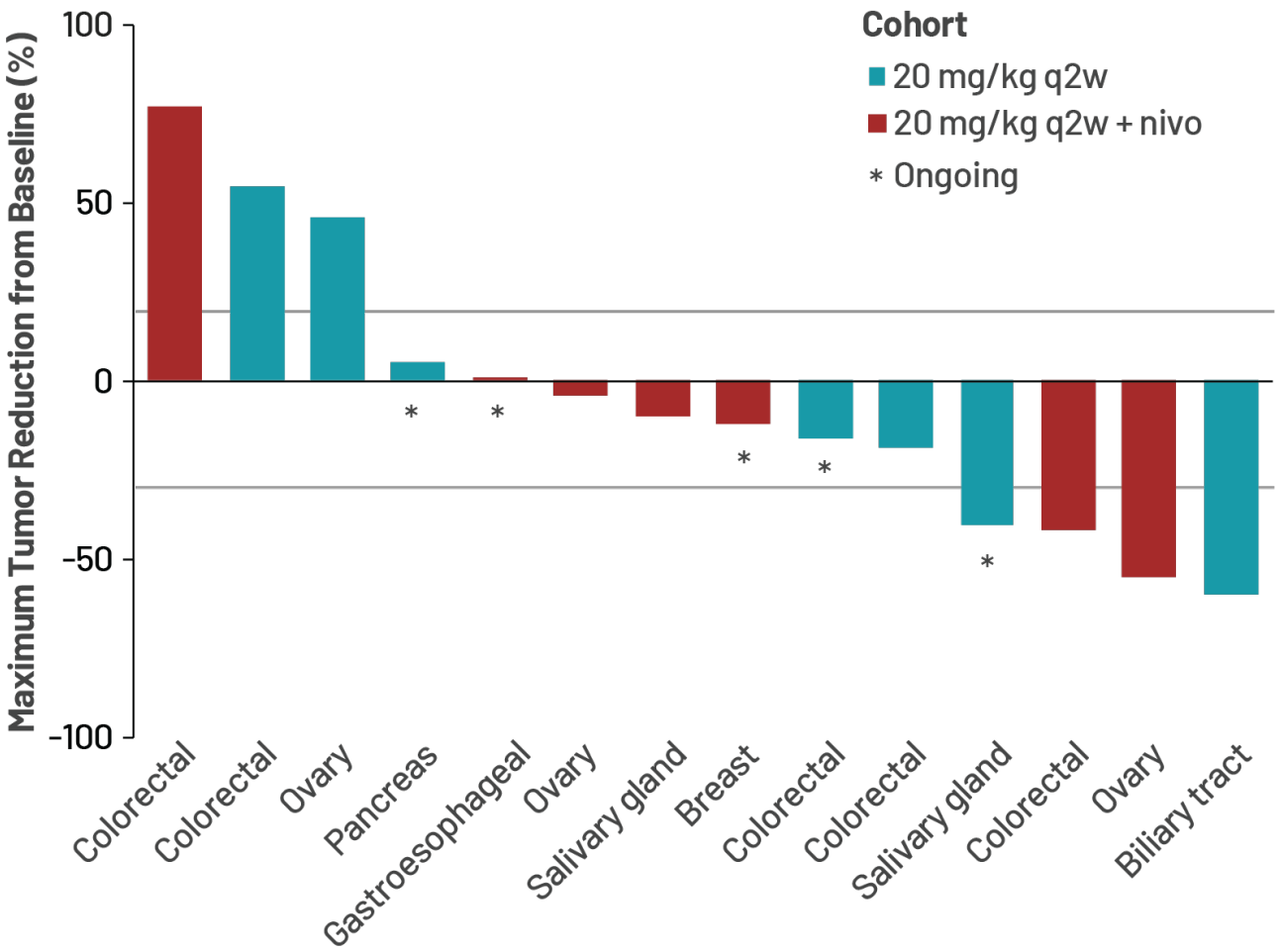
- Intravenous administration
- Local activation of the innate immune system
- Generates a durable tumor-targeted adaptive immune response






Ackerman SE, et al. *Nature Cancer*. 2021;2(1):18-33.

# Meaningful Anti-tumor Activity in Evaluable Heterogeneous (8 tumor types) HER2+ Tumor Population at the RP2D = 20 mg/kg q2w BDC-1001 Monotherapy and Combination with Nivolumab




- HER2+ either assessed by protein or gene analysis determined at enrollment
- RECIST v1.1 assessment criteria

## Monotherapy (n=7)

- 29% achieved PR
- 43% had disease control ≥24w
- 57% achieved tumor shrinkage
  - Tumor types: colorectal, salivary gland, and biliary tract

## Combination with Nivolumab (n=7)

- 29% achieved PR
- 57% (N=12) had disease control ≥24w
- 71% achieved tumor shrinkage
  - Tumor types: breast, colorectal, ovary, and salivary gland

Data cut-off: March 24, 2023

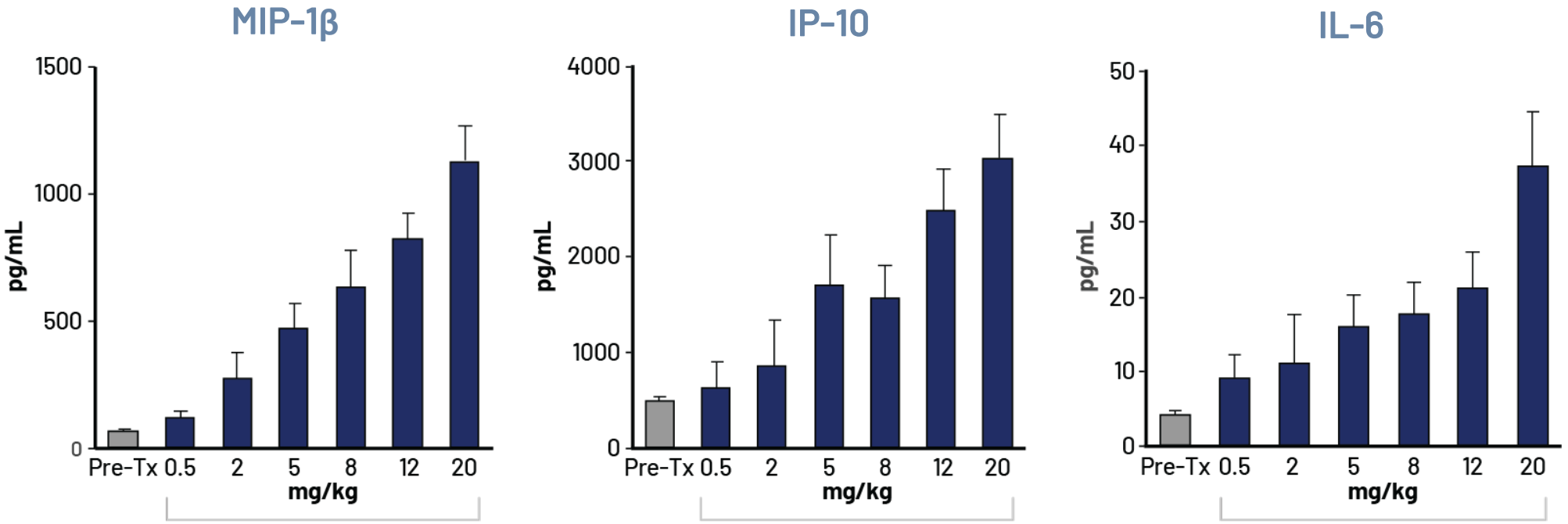


Bob T. Li, Mark D. Pegram, Keun-Wook Lee, et al. J Clin Oncol 41, 2023 (suppl 16; abstr 2538).

# Peak Increase in Plasma Myeloid Activation Markers at 4 Hours
## Confirms MOA and Safety Profile

- Plasma samples for cytokines and chemokines obtained from all patients
- Dose-dependent peak increases in Cycle 1 were observed in multiple cytokines and chemokines*
  - Similar responses observed for MIP-1α, IFNγ, TNFα and eotaxin

Average IL-6 levels were low at all doses (< 50 pg/mL)




Data cut-off: March 24, 2023

*Representative graphs are shown



Bob T. Li, Mark D. Pegram, Keun-Wook Lee, et al. J Clin Oncol 41, 2023 (suppl 16; abstr 2538).

# Details of Safety Profile of BDC-1001 Monotherapy and in Combination with Nivolumab

## Summary of Treatment-related TEAEs

| | BDC-1001 Monotherapy | | | | BDC-1001 + Nivolumab | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Treatment-related TEAEs | | | | BDC-1001 Treatment-related TEAEs | | | BDC-1001 + Nivolumab Treatment-related TEAEs | | |
| | q3w n = 52 | q2w n = 22 | q1w n = 20 | Total n = 94 | q2w n = 17 | q1w n = 20 | Total n = 37 | q2w n = 17 | q1w n = 20 | Total n = 37 |
| All grades (%) | 30 (57.7) | 11 (50.0) | 17 (85.0) | 58 (61.7) | 11 (64.7) | 14 (70.0) | 25 (67.6) | 5 (29.4) | 12 (60.0) | 17 (45.9) |
| Grade ≥3 (%) | 5 (9.6) | 1 (4.5) | 1 (5.0) | 7 (7.4) | 0 | 2 (10.0) | 2 (5.4) | 0 | 1 (5.0) | 1 (2.7) |
| Serious adverse events (%) | 3 (5.8) | 0 | 0 | 3 (3.2) | 1 (5.9) | 1 (5.0) | 2 (5.4) | 0 | 1 (5.0) | 1 (2.7) |
| Leading to treatment discontinuation | 3 (5.8) | 1 (4.5) | 0 | 4 (4.3) | 0 | 1 (5.0) | 1 (2.7) | 0 | 1 (5.0) | 1 (2.7) |
| Leading to treatment interruption | 5 (9.6) | 2 (9.1) | 2 (10.0) | 9 (9.6) | 1 (5.9) | 1 (5.0) | 2 (5.4) | 0 | 1 (5.0) | 1 (2.7) |
| Leading to death | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

Data cut-off: March 24, 2023

Safety graded by CTCAE v5; TEAE, treatment-emergent adverse event
Definition of treatment-related TEAEs = an AE considered as related to with unknown/missing relationship to study drug



Bob T. Li, Mark D. Pegram, Keun-Wook Lee, et al. J Clin Oncol 41, 2023 (suppl 16; abstr 2538).

# BDC-1001 Summary:

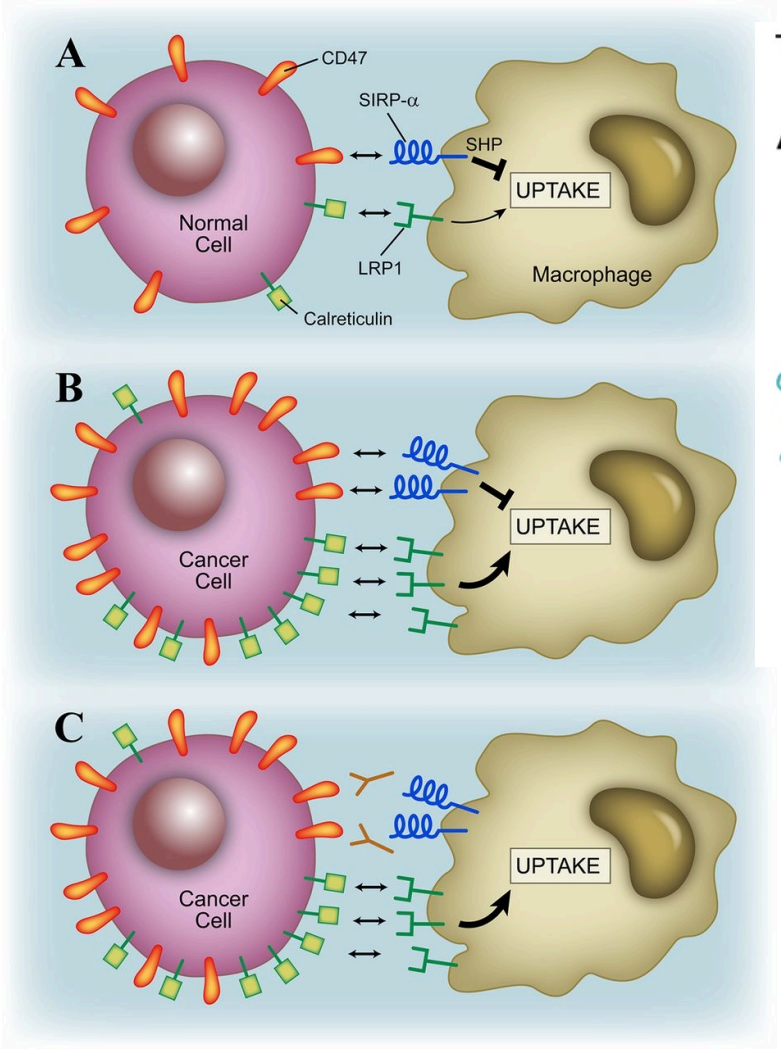
Results demonstrate encouraging evidence of safety, anti-tumor efficacy, and biomarker changes consistent with MoA of ISAC technology

- BDC-1001 was well-tolerated at all doses and dosing frequencies up to 20 mg/kg q1w
  - In a heterogeneous (16 different tumor types in 18 cohorts) and heavily pretreated (median 4 prior lines of systemic treatment) patient population
- Target exposure established in preclinical models achieved at higher dose and increased frequency of administration
  - $C_{min}$ above 10 µg/mL achieved at q2w and q1w schedules
  - Improved efficacy observed with q2w compared to q3w or q1w
- Clinical activity of BDC-1001 observed alone and in combination with nivolumab, particularly in the 20 mg/kg q2w cohorts
- Pharmacodynamic responses in both plasma and tissue consistent with ISAC MOA
  - Responses of myeloid and T cell activation and infiltration not anticipated with trastuzumab treatment alone
- Selection of 20 mg/kg q2w as RP2D based on the totality of safety, efficacy, PK, and biomarkers
- Results support Phase 2 development of BDC-1001 as a single agent and in combination strategies

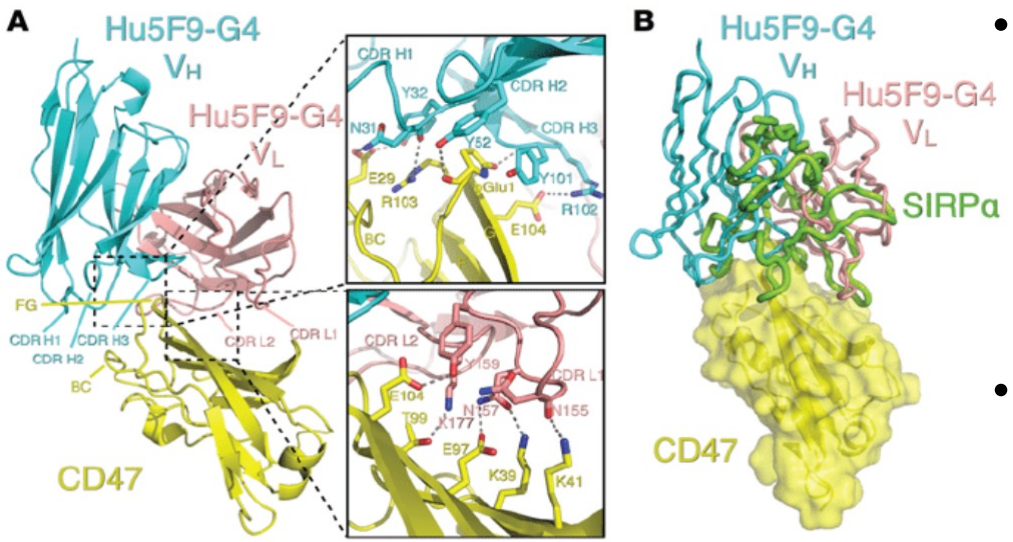


Bob T. Li, Mark D. Pegram, Keun-Wook Lee, et al. J Clin Oncol 41, 2023 (suppl 16; abstr 2538).

# Macrophages ignore CD47+ cells as a result of negative interactions in which the CD47–SIRP-α pair promote a “don’t eat me” signal; humanized CD47 antibody blocks SIRPα interaction




The Journal of Clinical Investigation



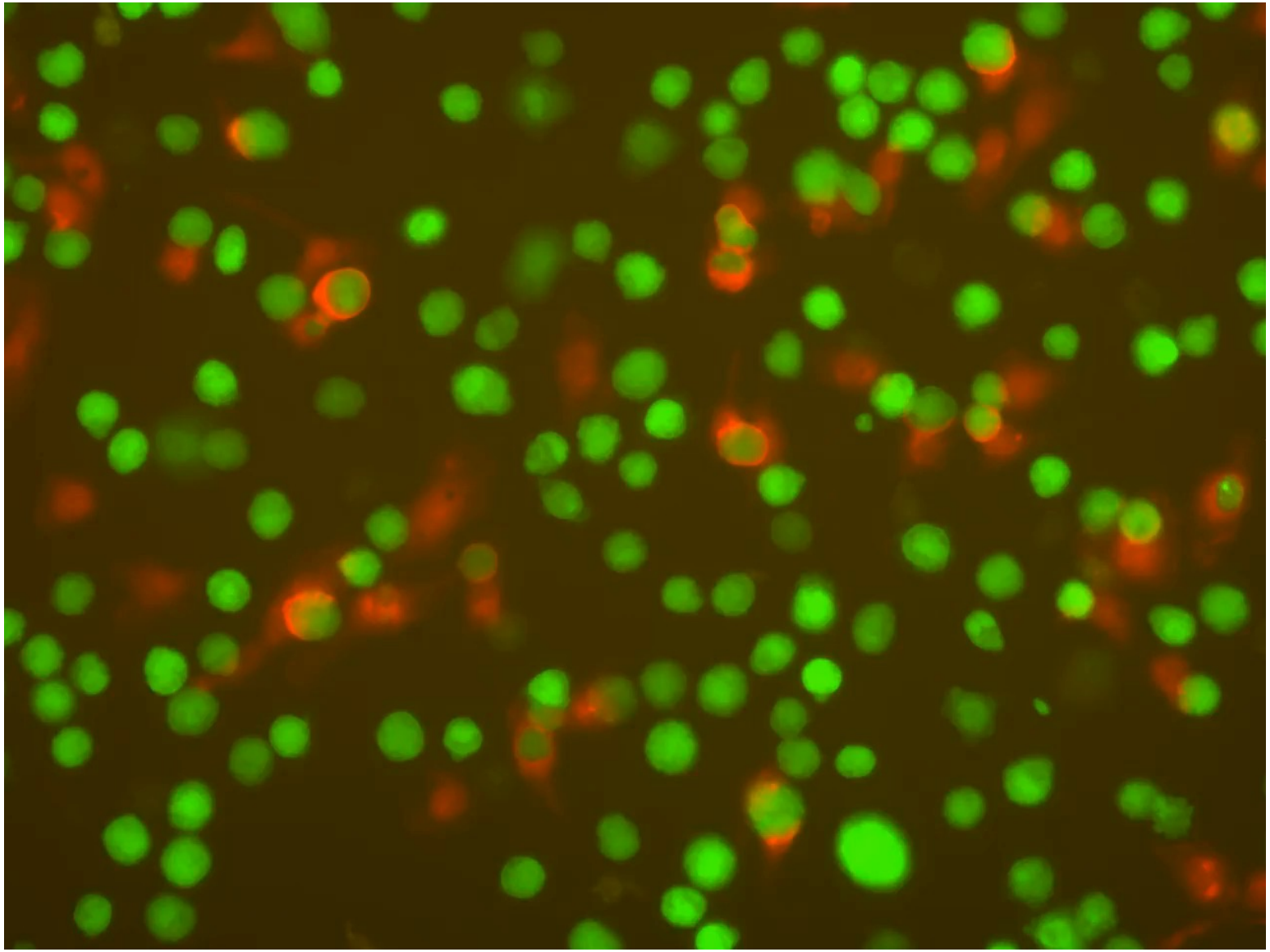

Crystal structure of CD47-ECD in complex with Hu5F9-G4: A) Hu5F9-G4/CD47 interface; B) Superposition of SIRPα demonstrating a shared binding interface.

- Hu5F9-G4 (magrolimab) binds human CD47 with high affinity by Biacore:
  8-10 nM for monomeric CD47
  8 pM for bivalent CD47
- Engineered into human IgG4 Fc (to avoid ADCC/CDC), with Ser-Pro substitution to reduce Fab arm exchange
- Humanized by CDR grafting

**Weiskopf K, et al. J Clin Invest. 2016;126(7):2610-2620.**

**Unanue ER PNAS 2013;110:10886-10887.**

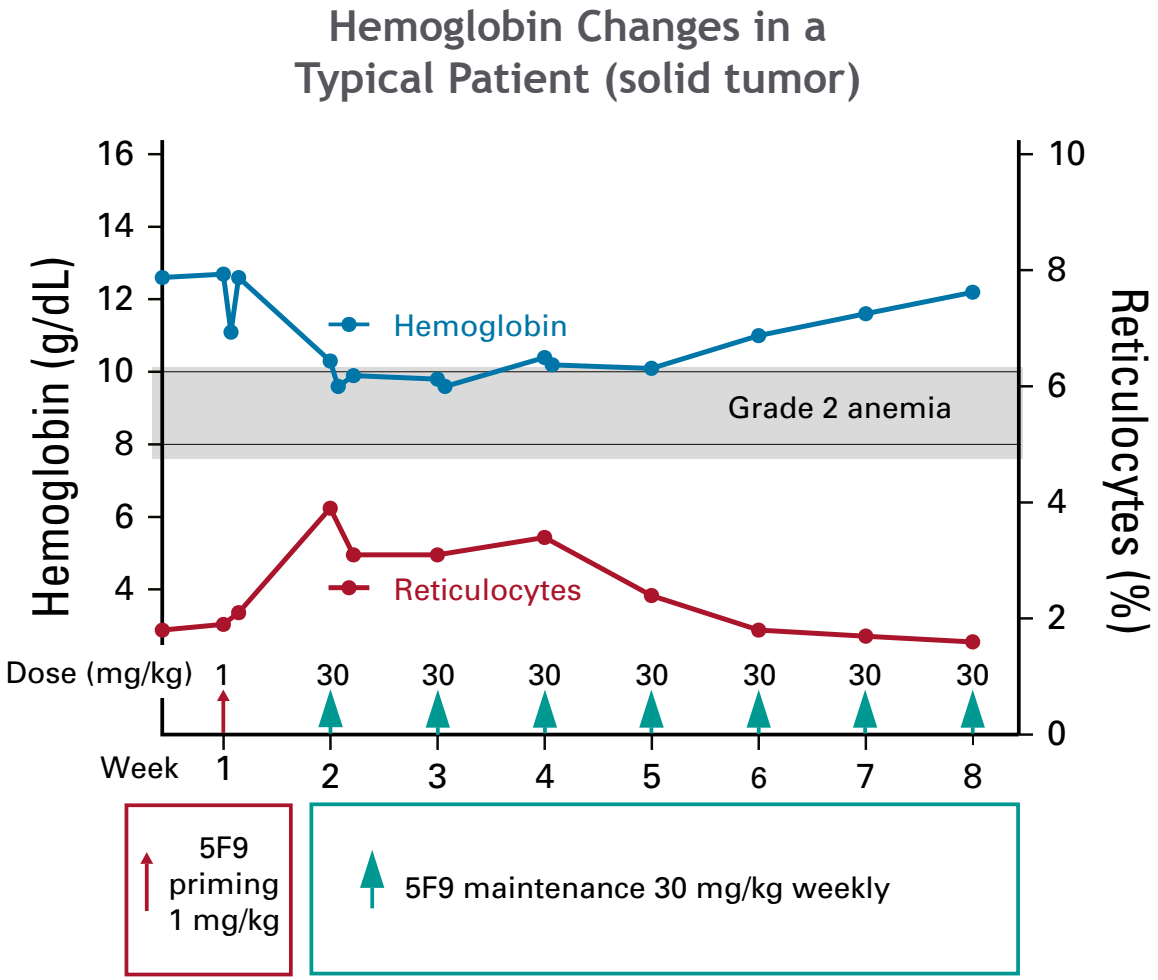
# Macrophages (red) phagocytosing CD47+ tumor cells (green) in the presence of anti-CD47 antibody



MΦ -- red
CD47+ tumor cells -- green

Edris, B. et al.. PNAS 109, 6656–6661 (2012).

# On Target Anemia is a Pharmacodynamic Effect and is Mitigated with a Priming and Maintenance Dosing Regimen




Hemoglobin lower limit of normal > 11.7-13.5 g/dL
Reticulocytes upper limit of normal < 2.28%

**An example patient on the Phase 1 solid tumor study of magrolimab evaluating safety and dose regimen**

- Aging RBCs can be cleared by CD47 blockade leading to an on-target anemia
- Initial priming dose of 1 mg/kg results in a temporary and mild decline in hemoglobin through clearance of aged RBCs and a temporary reticulocytosis that resolves
- Hemoglobin levels return to baseline even with continued treatment with magrolimab at significantly higher doses (30 or 45 mg/kg)
- Average hemoglobin drop with the first priming dose was mild (0.8 g/dL) across patients -- 4/62 pts had PRBC transfusion

Sikic et al., J Clin Oncol. 2019;37(12):946-953.

# Confirmed Objective Partial Responses to Single Agent Hu5F9-G4

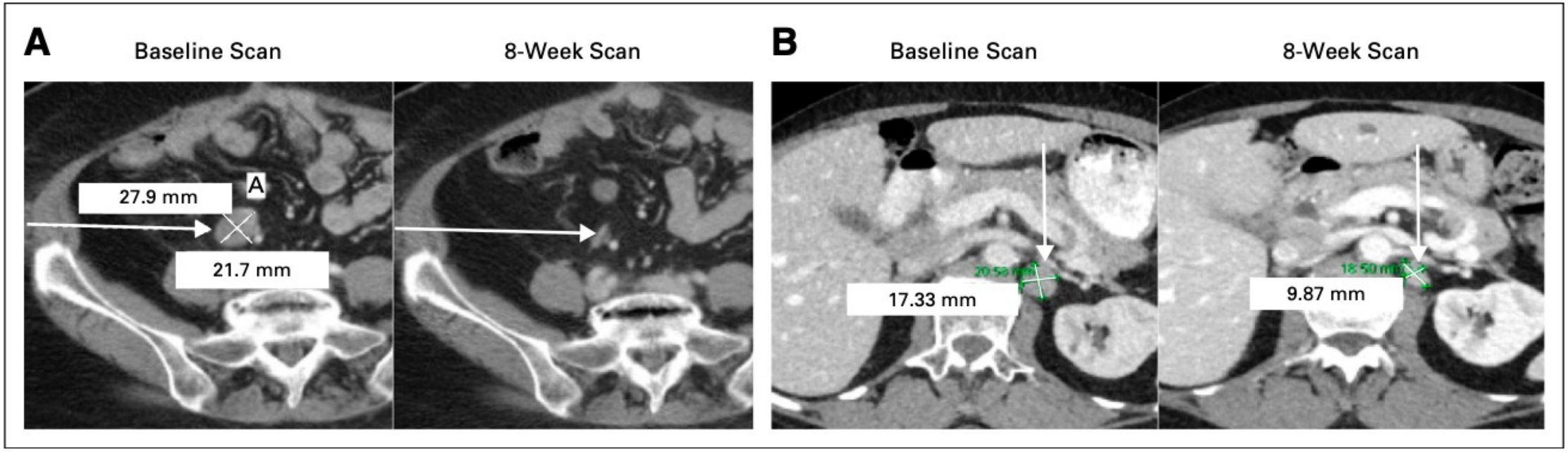



**FIG 3.** Hu5F9-G4 partial responses observed in (A) a patient with clear cell ovarian cancer and (B) a patient with fallopian tube cancer.

Time to progression of 5.2 months (A) and 9.2 months (B), respectively. CA125 levels dropped from 338 to 70 U/mL and 890 to 103 U/mL, respectively. Both patients were heavily pretreated with more than six prior lines of systemic therapy, and both received 5F9 at 20 mg/kg.

**Non-linear PK at low doses due to CD47 antigen sink (erythrocytes, etc.); PD – 100% receptor occupancy at 30mg/kg; [trough] ≥ 200ug/mL; RP2D = 30mg/kg q 2 weeks.**

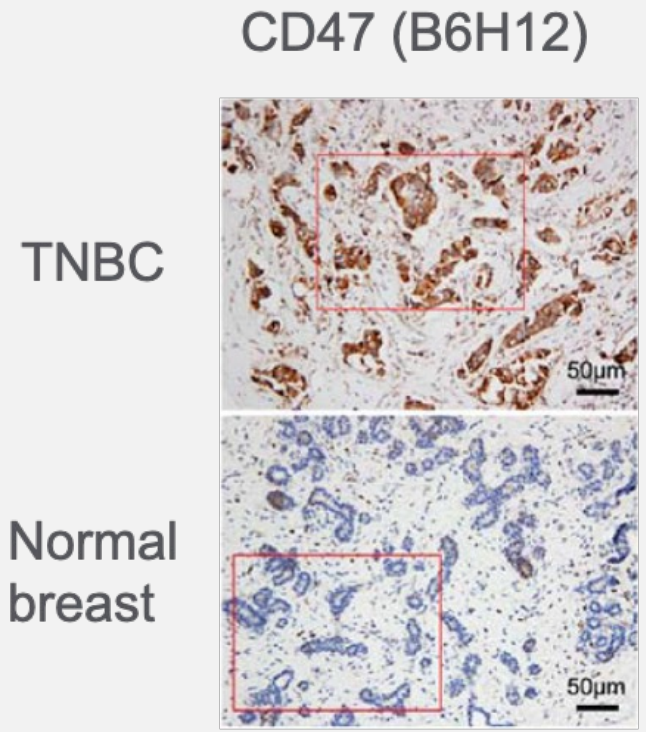
AEs of interest – anemia, hemagglutination (no clinical sequelae) and IRRs.

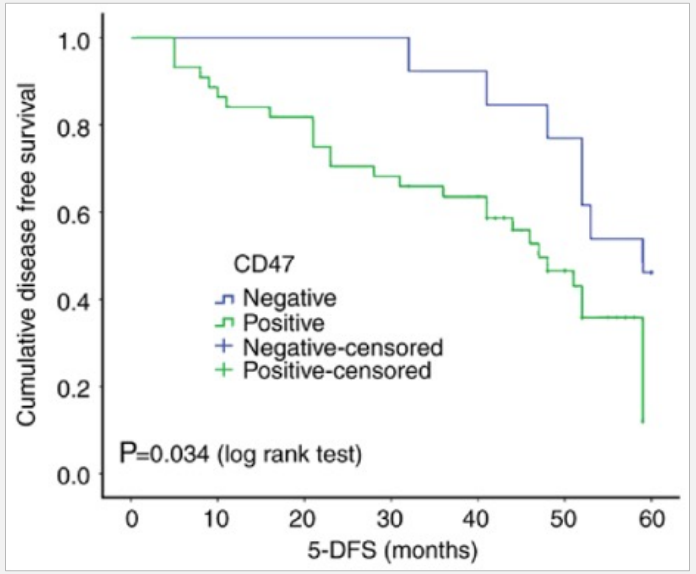
Stanford University

Sikic et al., J Clin Oncol. 2019;37(12):946-953.

# CD47 expression is associated with poor prognosis in triple negative breast cancer

- CD47 is detected by IHC (clone B6H12) in >75% of TNBC cases
- Expression is more prevalent in advanced disease, including higher TNM stage, presence of LN metastases, and recurrent disease
- CD47 positive TNBC cases have reduced disease free survival

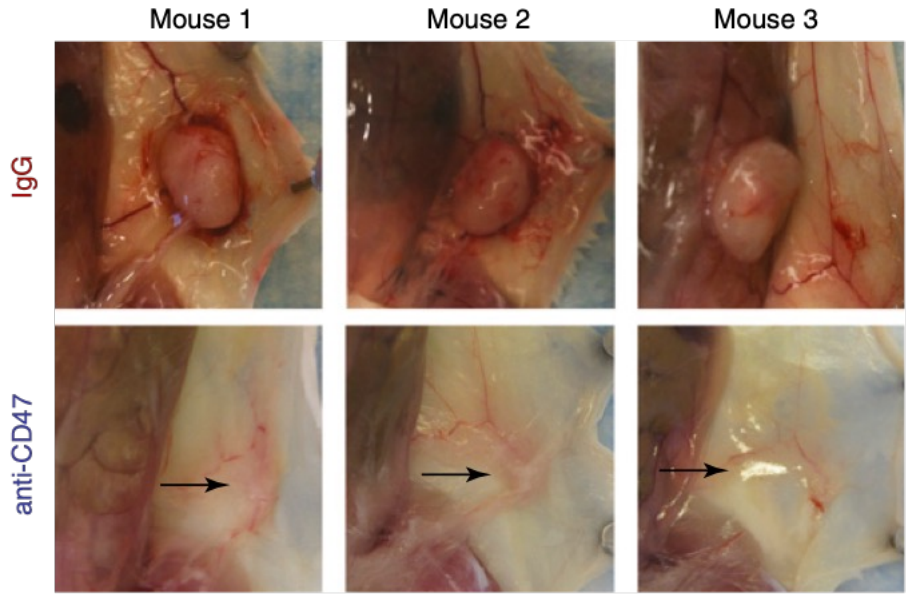


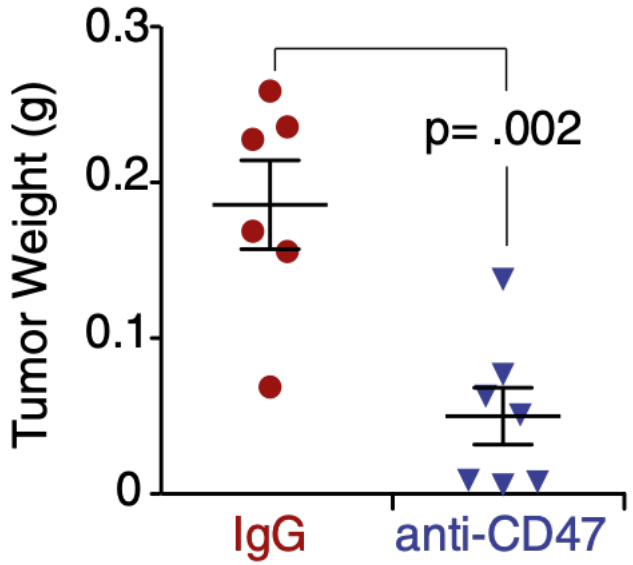




| Groups | All cases, n | CD47 protein expression | | |
|---|---|---|---|---|
| | | Positive, n (%) | $\chi^2$ | P-value |
| Breast benign lesions | 40 | 12 (30.0) | 21.453 | <0.001 |
| Triple-negative breast cancer tissues | 57 | 44 (77.2) | | |

*Yuan (2019) Oncol Lett 18:3249.*

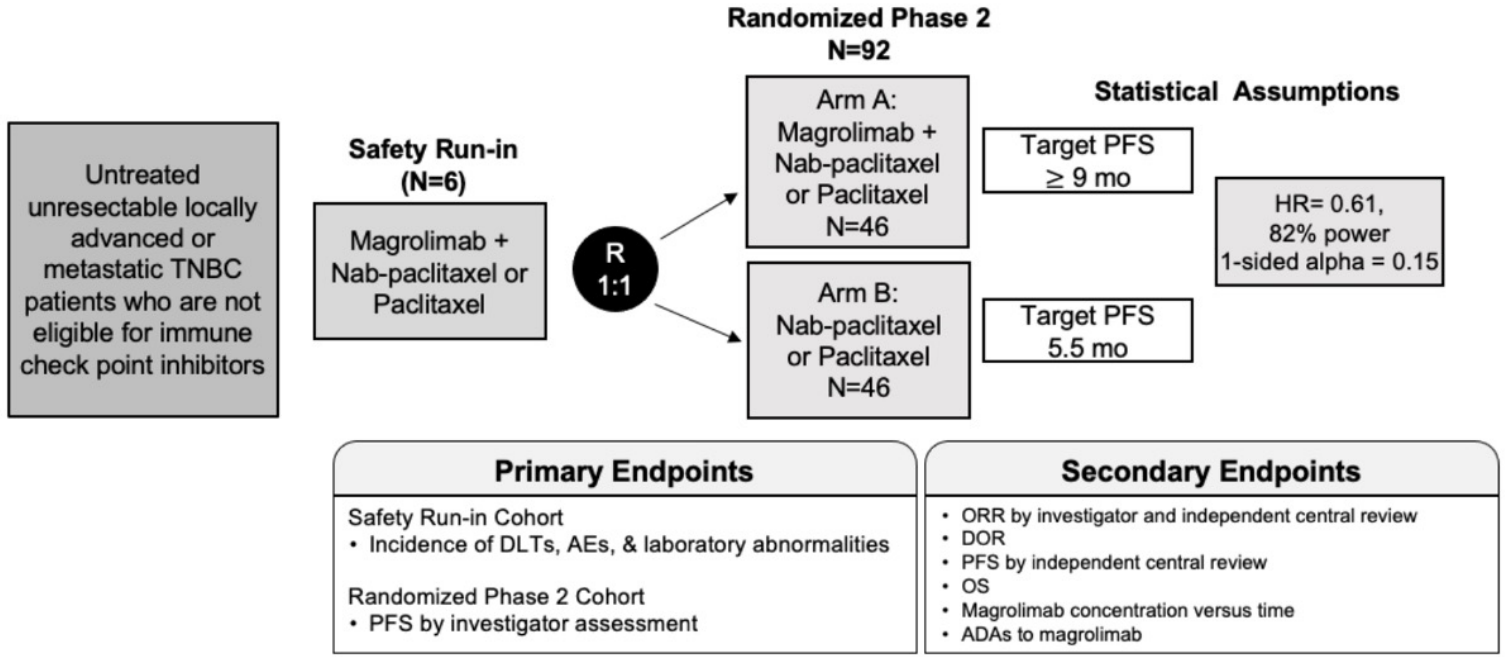
# Anti-CD47 Antibody Significantly Reduces TNBC PDXs *in vivo* → Clinical Translation: Randomized Phase II




Willingham et al., PNAS 109 (42) E2842.

## Study Schema




| Primary Endpoints | Secondary Endpoints |
|---|---|
| Safety Run-in Cohort<br>• Incidence of DLTs, AEs, & laboratory abnormalities<br><br>Randomized Phase 2 Cohort<br>• PFS by investigator assessment | • ORR by investigator and independent central review<br>• DOR<br>• PFS by independent central review<br>• OS<br>• Magrolimab concentration versus time<br>• ADAs to magrolimab |

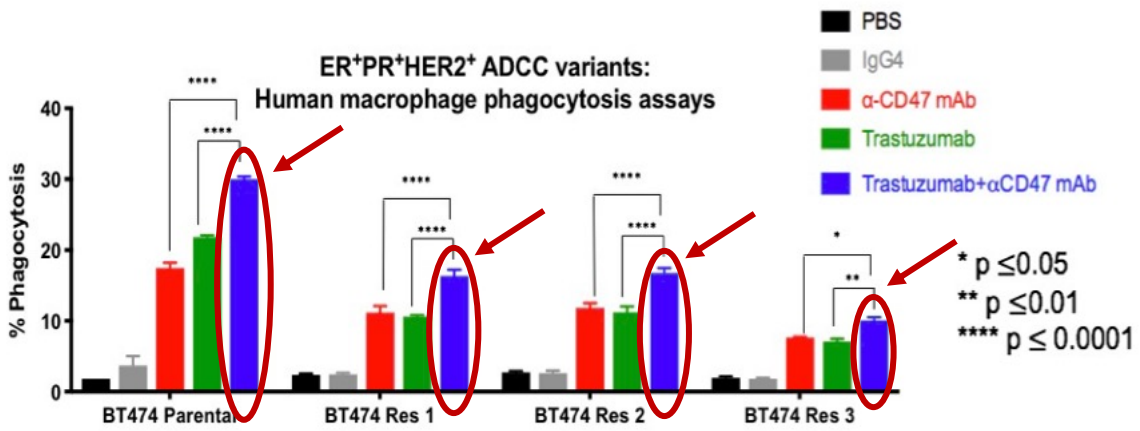
ADA = anti-drug antibodies; AE = adverse event; DOR = duration of response; HR = hazard ratio; ORR = objective response rate; OS = overall survival; PFS = progression-free survival; R = ratio; TNBC = triple-negative breast cancer

- CD47 is expressed on TNBC-derived cell lines
- CD47 blockade enhances TNBC phagocytosis
- Anti-CD47 dependent phagocytosis is further augmented by Paclitaxel
- 2nd cohort - Sacituzumab govitecan ± magrolimab

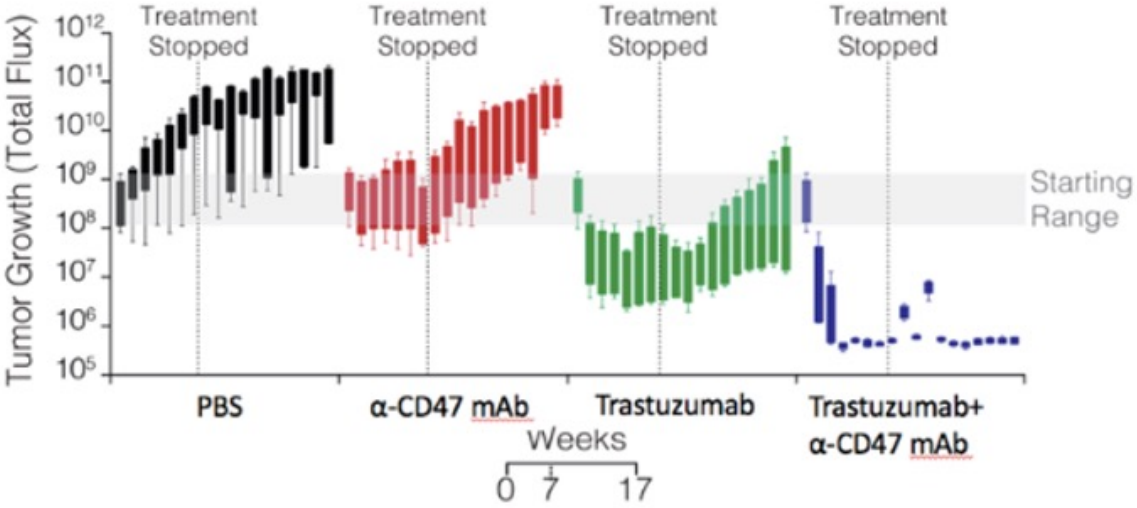
PNAS
Proceedings of the National Academy of Sciences of the United States of America www.pnas.org

# MOA – ADCP: Combining CD47 blockade with trastuzumab eliminates HER2-positive breast cancer cells to overcome trastuzumab ADCC tolerance

***In vitro:***
***The circled blue bars (red arrows) indicate HER2+ ADCC-tolerant breast cancer cells being phagocytosed by human macrophages***



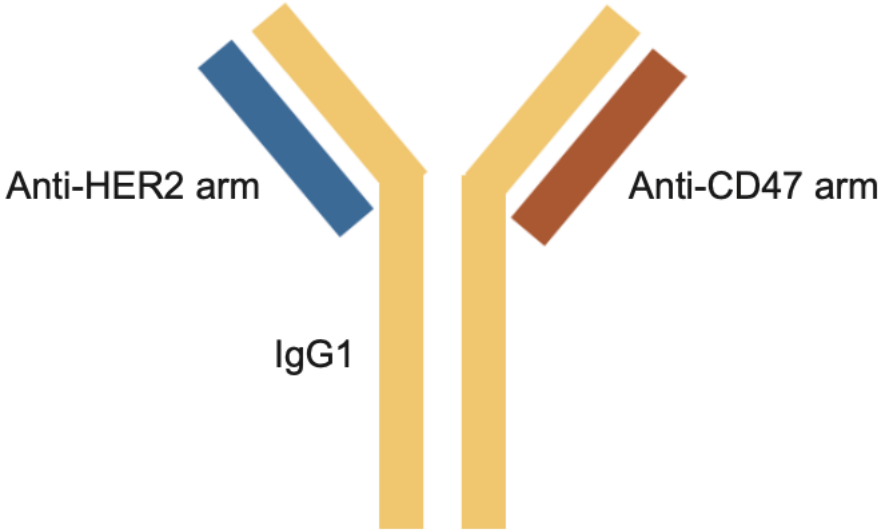

***In vivo:***
***Greater combined efficacy of anti-CD47 antibody magrolimab plus trastuzumab against HER2+ human GFP-luciferase BT474 breast cancer xenografts***



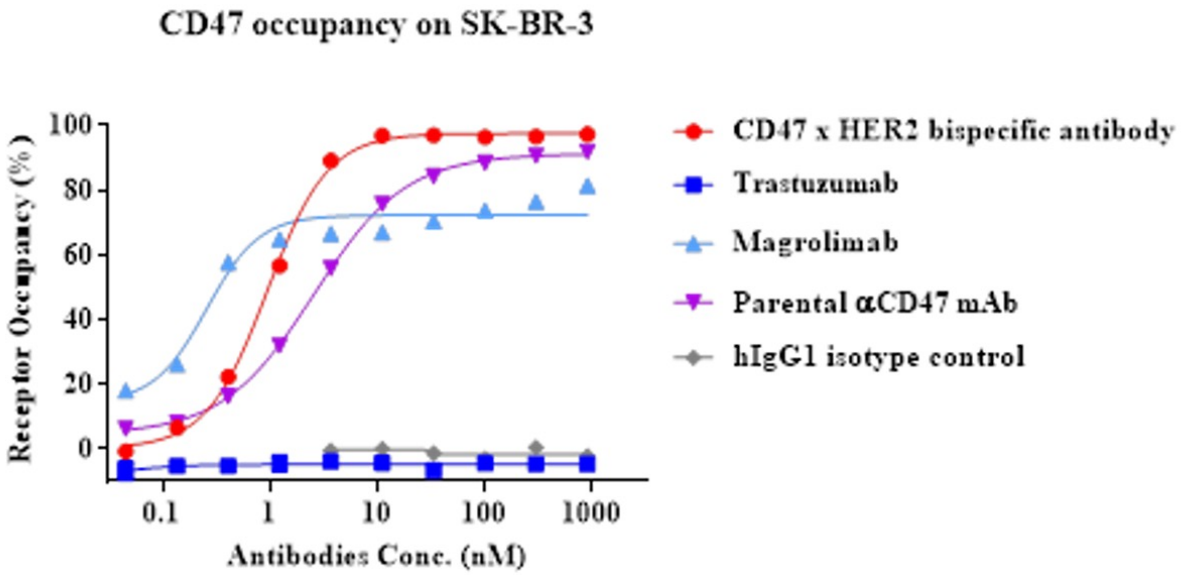

Tumor burden was measured using *in vivo* bioluminescence imaging

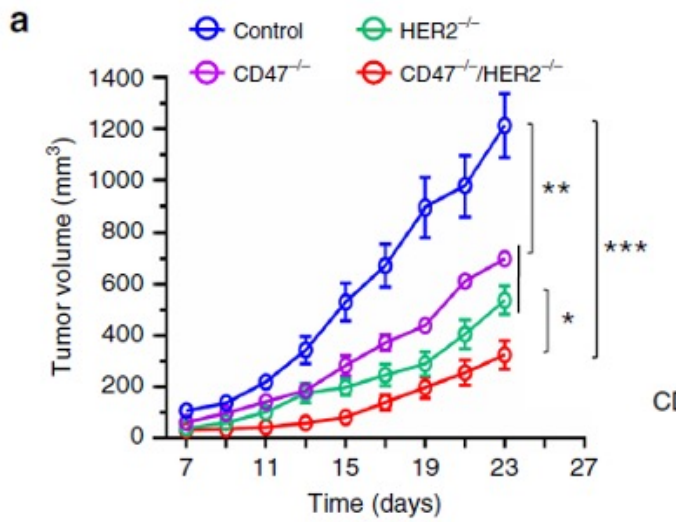
Rosalynd Upton,…Mark D. Pegram[1], and Irving L. Weissman[1]. Proceedings of the National Academy of Sciences Jul 2021, 118 (29) e2026849118. [1]M.D.P. and I.L.W. contributed equally to this work.

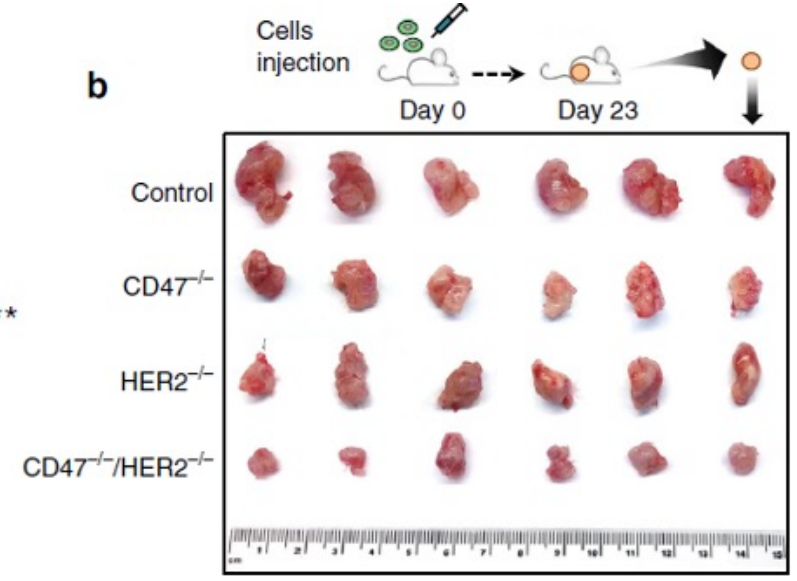
# D3L-001 Bispecific HER2 X CD47 MAb

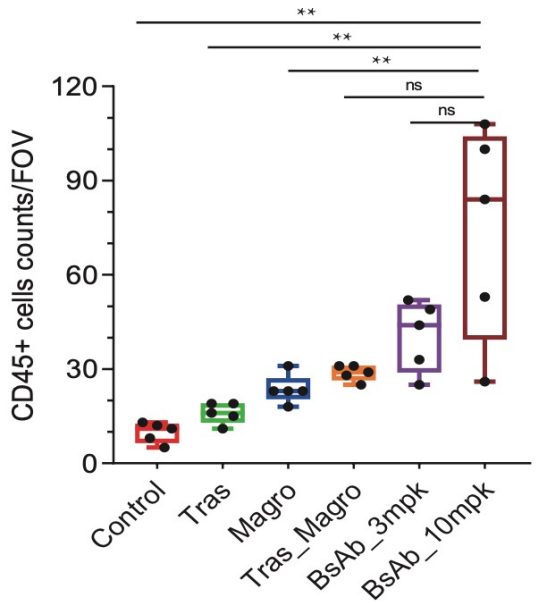



**Occupancy on SK-BR-3: BsAb EC50=1 nM; Parental CD47 EC50= 3 nM**



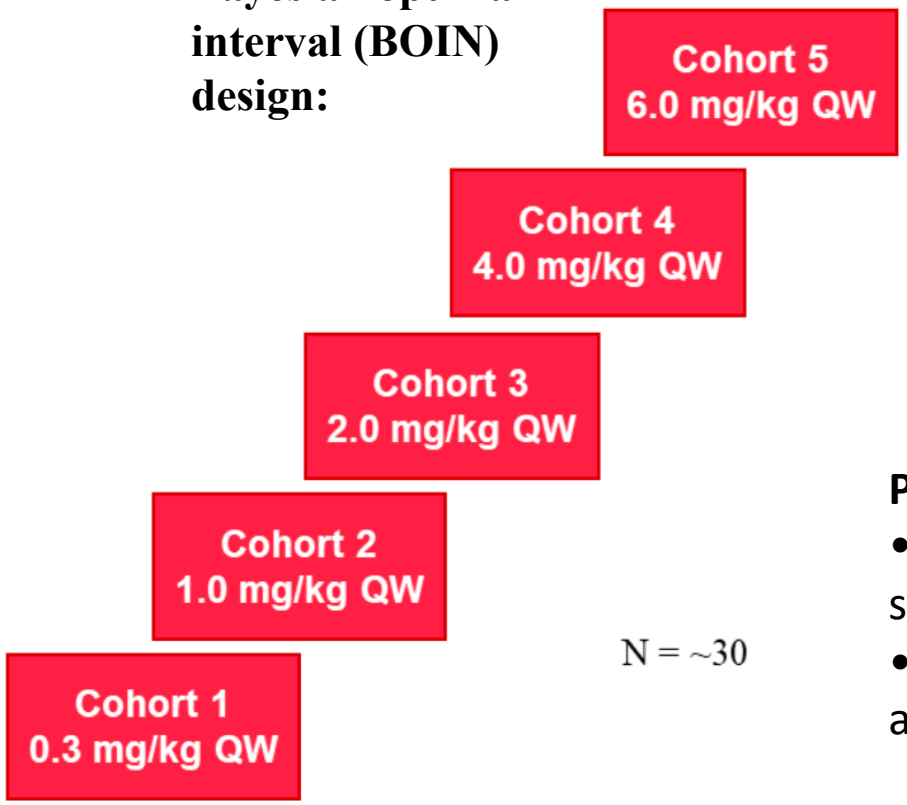



## 1.2. Schema

**Part 1 – Dose Escalation**

**Bayesian optimal interval (BOIN) design:**




MTD and RP2D, if applicable

Recommended dose for expansion

**Part 2 – Dose Expansion**

Cohort A, 3L+HER2-positive breast cancer, N=~40

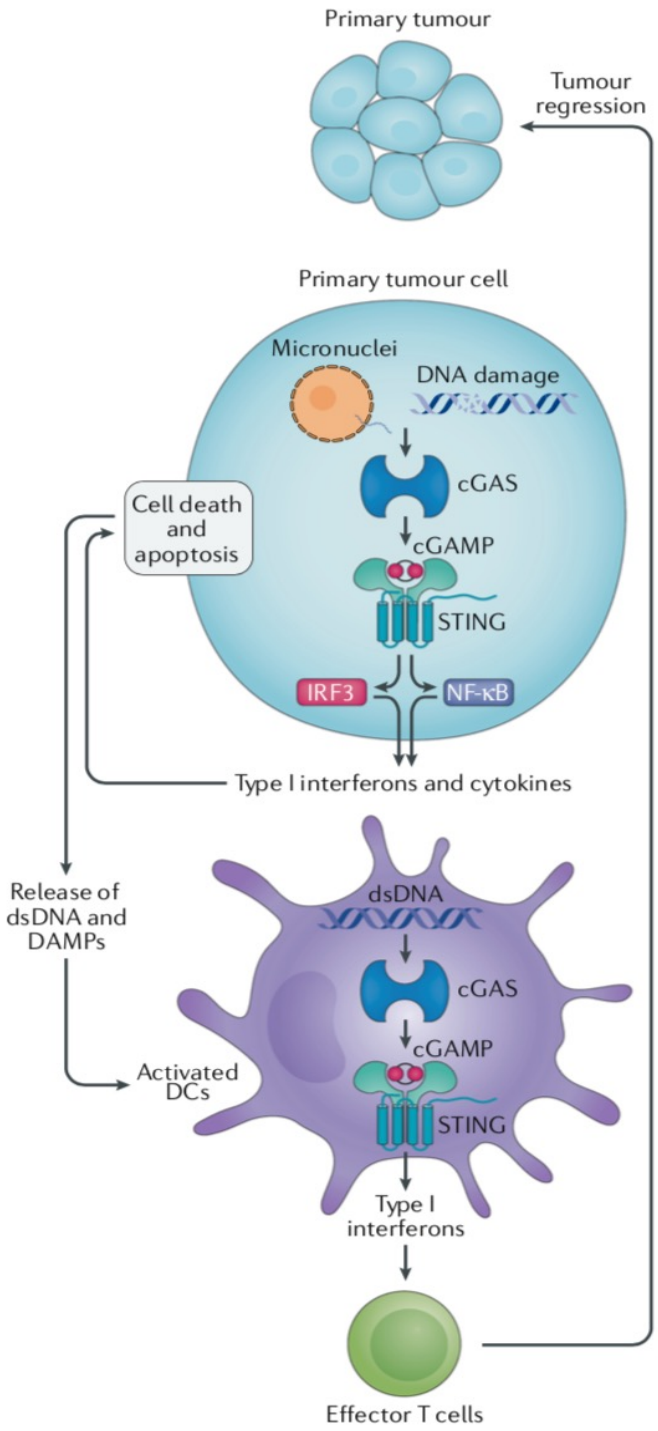
Cohort B, 3L+HER2-positive gastric/gastroesophageal junction adenocarcinoma, N=~40

**Primary Objectives:**

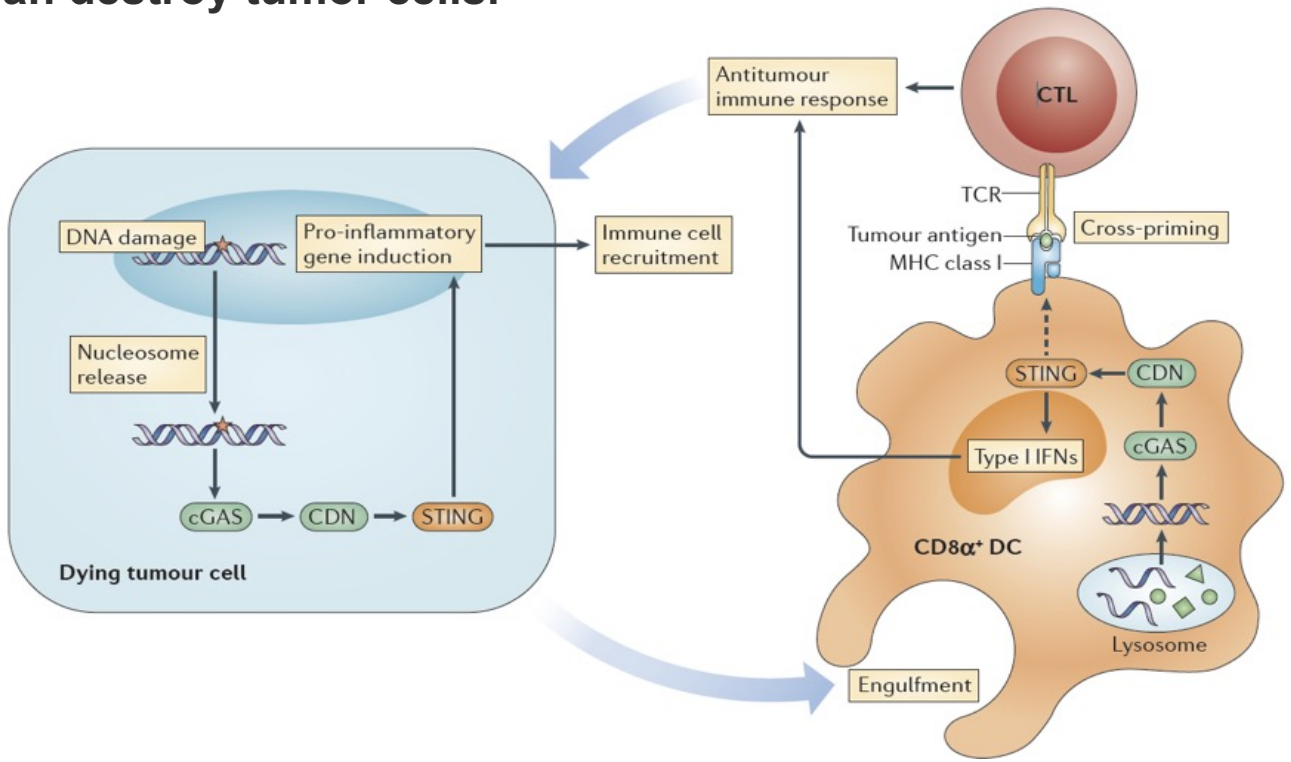
- To evaluate the safety and tolerability of D3L-001 in adult subjects with HER2-positive solid tumors.
- To determine the maximum tolerated dose (MTD), if applicable, and the recommended phase 2 dose (RP2D).

Abbreviations: HER2, human epidermal growth factor receptor 2; MTD, maximum tolerated dose; N, number of subjects; QW, once weekly; RP2D, recommended phase 2 dose; SRC, safety review committee.
In addition to current dose levels in the schema, based on the review and recommendation by the SRC, intermediate dose level(s), de-escalation to below 0.3 mg/kg QW (eg, 0.1 mg/kg QW) and escalation to beyond 6.0 mg/kg QW, may be explored. For escalation to beyond 6.0 mg/kg QW, increment of dose level would be no more than 50%.

# STING: an essential link between innate and adaptive immunity is provided by dendritic cells

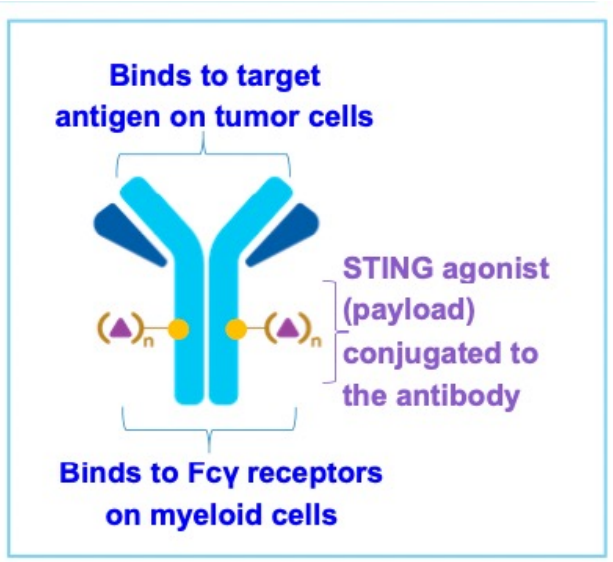


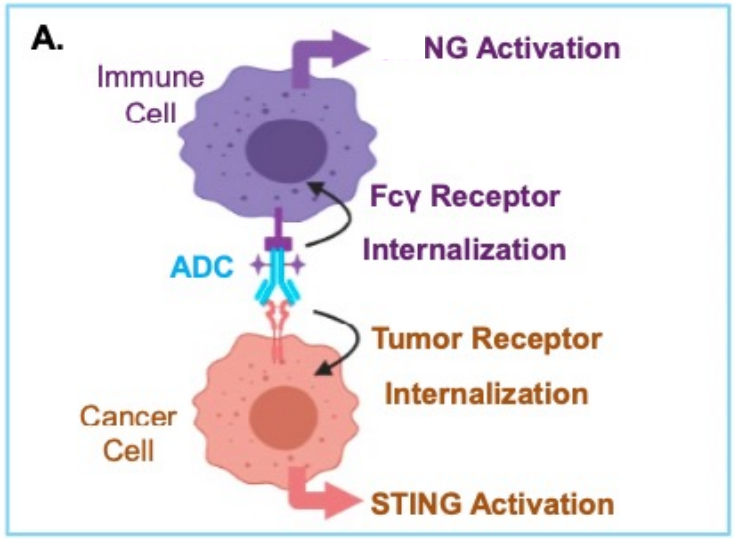

**Uptake of dsDNA and tumor antigens by tumor-resident dendritic cells (DCs) elicits a complimentary cGAS–STING-dependent type I interferon-mediated activation of an antitumor immune response, for example, through activation of effector T cells such as tumor-associated, antigen-specific CD8+ T cells, which can destroy tumor cells.**

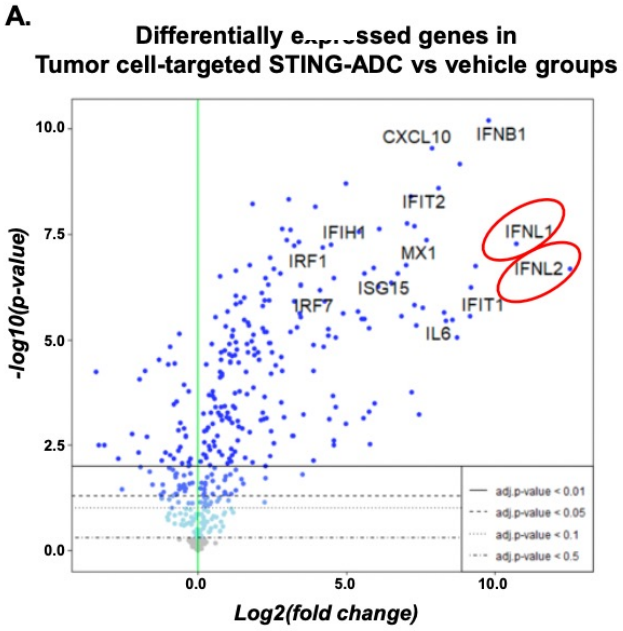


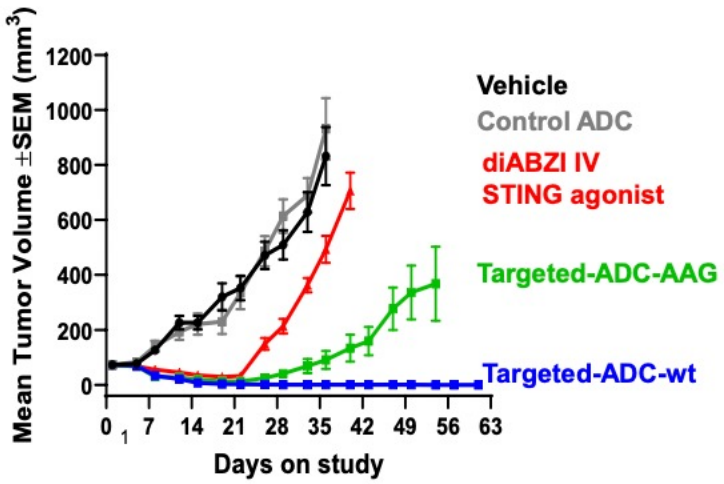

Motwani M, Pesiridis S, Fitzgerald KA.
Nat Rev Genet. 2019 Nov;20(11):657-674.

# Tumor cell-intrinsic STING pathway activation leads to robust induction of Type III Interferons and contributes to the anti-tumor activity elicited by STING agonism



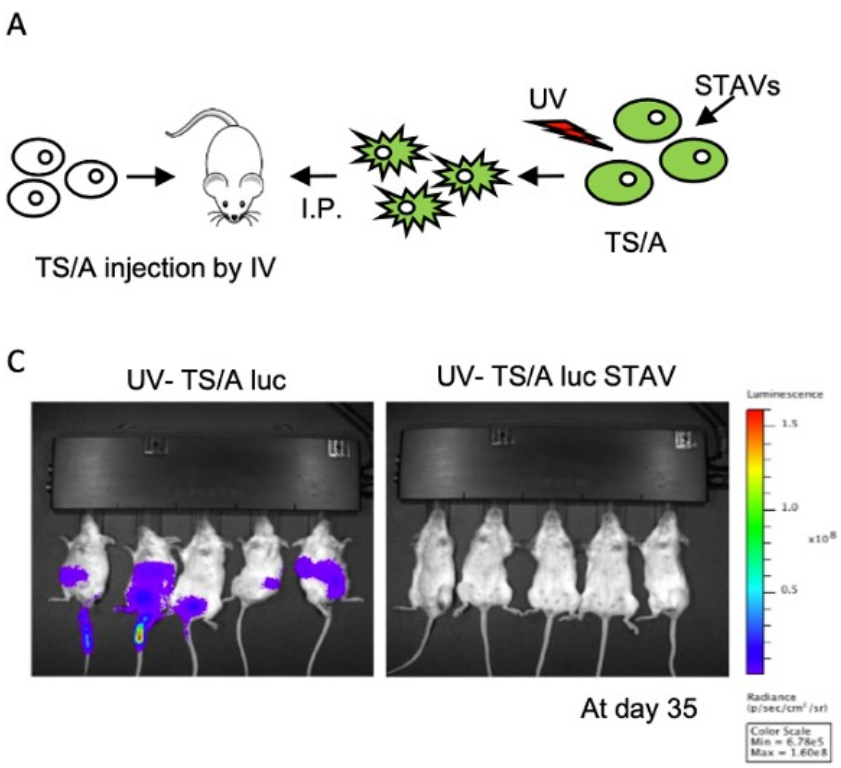



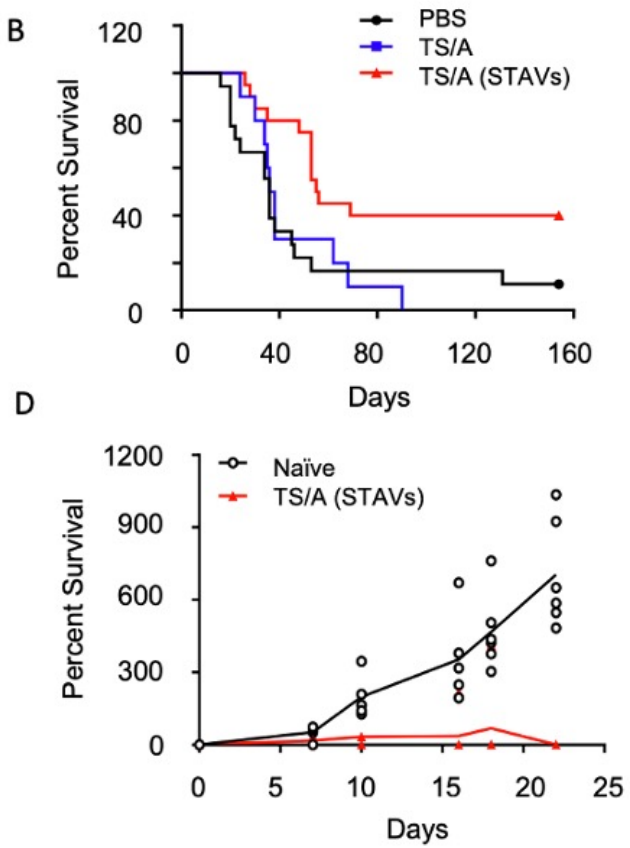

March 13, 2023 Press Release:

Clinical hold by FDA placed on XMT-2056 phase I clinical trial, due to recent grade 5 serious adverse event deemed to be drug-related. The SAE and its cause remain under investigation.

Cetinbas NM, et al. Proceedings of the American Association for Cancer Research 2021: abstr 1773.

# STing-Activating adjuVants (STAVs) are an effective cell-based therapy for breast cancer



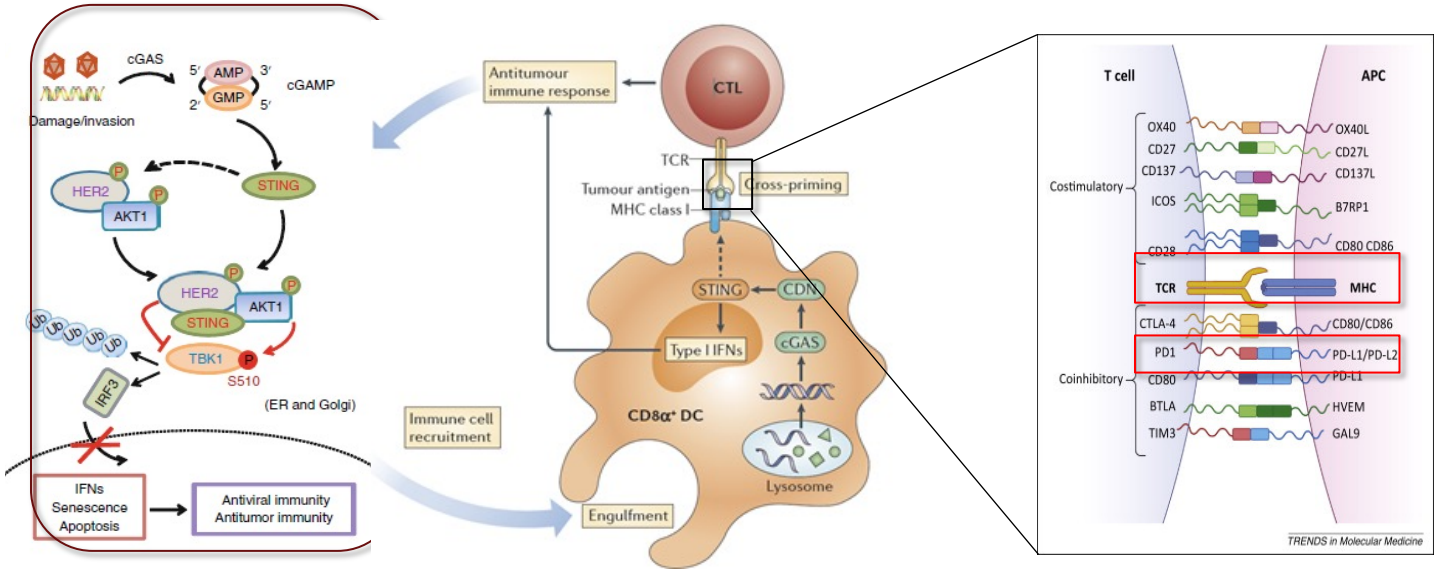




**Effect of UV-irradiated, STAV-treated TS/A breast carcinoma cells in IV TS/A lung metastasis model.** *(A) Schematic representation of experimental design; (B) TS/A (STAVs)-treated, UV-irradiated cells prolong survival (N=20/group, p=0.008); (C) Day 35 luciferase activity in UV-TS/A (luc) STAV-treated (right) mice as compared control-treated mice (left); (D) TS/A-luc STAV-treated mice rechallenged with TS/A-luc cells (1X10$^5$) vs. naïve mice controls (6/group; p=0.03).*



Ahn et al., 2018, Cancer Cell 33, 862–873.

# Reconstituting cGAS-STING signaling in HER2+ breast cancer: Summary




1. The detection of pathogens through nucleic acid sensors is a defining principle of innate immunity. DNA-sensing receptors sample subcellular compartments for foreign nucleic acids and, upon recognition, trigger immune signaling pathways for host defense.
2. Aberrant DNA fragments are ubiquitous in cancer cells due to abnormal chromosome structure, genome instability and post-radiation/chemo effects, which can be sensed by cGAS–STING. It is hypothesized evading damage surveillance is therefore necessary in tumorigenesis and tumor progression.
3. HER2 kinase inhibits cGAS–STING signaling, and prevents breast cancer cells from producing cytokines; cGAS-STING signaling may be reconstituted by anti-HER2 treatment.
4. Defects in STING signaling may enable HER2+ cells to escape cytokine production triggered by catastrophic DNA damaging events which would otherwise facilitate their eradication via the immune-surveillance system. A corollary to this hypothesis is that pharmacologic STING activation in concert with HER2 blockade will reconstitute the cGAS-STING innate immune signaling, setting the stage for therapeutic strategies aimed at amplifying effective adaptive anti-tumor responses.

STANFORD
CANCER INSTITUTE
Stanford University Medical Center
A NATIONAL CANCER INSTITUTE-DESIGNATED CANCER CENTER

# Questions/Comments
# Debate/Discussion
# Criticism

THANK YOU!

James H. Clark Center
Stanford University

Stanford Bio-X Program:
Biology, Medicine,Chemistry,
Physics and Engineering