The Bispecific Antibody Zanidatamab (ZW25) Has Unique Mechanisms of Action and Durable Anti-Tumor Activity in HER2-Expressing Cancers

Abstract: 1005

Nina Weisser[1], Grant Wickman[1], Libin Abraham[2], Jason O'Toole[1], Joy Guedia[1], Gesa Volkers[1], Joe Schrag[3], Charles Stevens[1], Kate Choi[2], Bryant Harbourne[1], Chi Wing Cheng[1], Peter Chan[1], Duncan Browman[1], Mike Gold[2], Neil Josephson[1], Surjit Dixit[1], Gerry Rowse[1]

[1] Zymeworks Inc., 1385 West 8th Ave, Vancouver, BC, Canada; [2] Department of Immunology and Microbiology, University of British Columbia, Vancouver, BC, Canada; [3] Human Health Therapeutics Portfolio, NRC-CNRC , Montreal, QC, Canada

Zanidatamab Monotherapy Shows Anti-Tumor Activity in Patients with Advanced HER2-Expressing Cancers

HER2-directed therapies have improved clinical outcomes for many patients with HER2-positive breast and gastric cancer. Despite these successes, there remains a need to develop improved HER2-targeted therapies for HER2-expressing tumors, particularly in the setting of recurrent or metastatic disease.

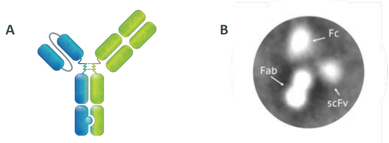
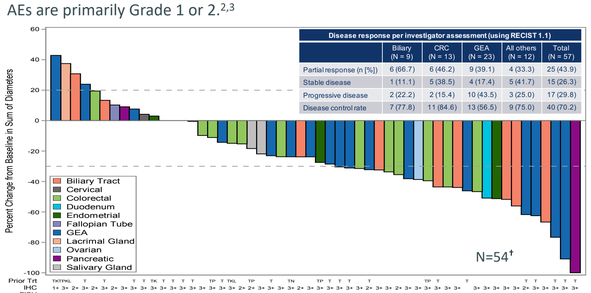


Figure 1. A) Zanidatamab is a humanized, bispecific, immunoglobulin (Ig) G1-like antibody (Ab) directed against the juxtamembrane extracellular domain (ECD4; scFv, blue) and the dimerization domain (ECD2; Fab, green) of HER2, the same domains targeted by trastuzumab (T; tras) and pertuzumab (P; pert) and is engineered with the Azymetric™ platform[1] B) Transmission electron microscopy showing the Fc, scFv and Fab domains of zanidatamab.

Zanidatamab is actively being evaluated in clinical trials in multiple HER2 expressing solid tumors. Studies to date show zanidatamab is well tolerated, treatment-related AEs are primarily Grade 1 or 2.[2,3]



Disease response per investigator assessment (using RECIST 1.1)	Biliary (N = 9)	CRC (N = 13)	GEA (N = 23)	All others (N = 12)	Total (N = 57)
Partial response (n [%])	6 (66.7)	6 (46.2)	9 (39.1)	4 (33.3)	25 (43.9)
Stable disease	1 (11.1)	5 (38.5)	4 (17.4)	5 (41.7)	15 (26.3)
Progressive disease	2 (22.2)	2 (15.4)	10 (43.5)	3 (25.0)	17 (29.8)
Disease control rate	7 (77.8)	11 (84.6)	13 (56.5)	9 (75.0)	40 (70.2)

Disease control rate defined as percentage of patients with complete response, partial response, or stable disease per RECIST 1.1
† 3 of the 57 response-evaluable patients had no post-baseline disease assessment of their target lesions and are not included in the waterfall. Response-evaluable includes all patients with measurable disease who had at least one post-baseline disease assessment (per RECIST 1.1) or discontinued study treatment prior to reassessment due to death from any cause or clinical progression (response imputed as Progressive disease). Colorectal cancer (CRC), gastroesophageal adenocarcinoma (GEA).

Figure 2. Zanidatamab has promising anti-tumor activity as monotherapy in patients with advanced HER2-expressing cancers that have progressed after standard of care therapies (median of 3 prior lines of therapy), including HER2-targeted agents trastuzumab (T), pertuzumab (P), T-DM1 (K), lapatinib (L) and neratinib (N)[3]. Results from ZW25-101 (NCT02892123), a first-in-human Phase 1 study that evaluates zanidatamab in HER2-expressing cancers. Data cutoff: Sept 18, 2019.

Phase 1 data support pivotal trial now enrolling in HER2 gene amplified biliary tract cancer (HERIZON-BTC-01; NCT04466891).

We have shown that zanidatamab's unique design and bispecific binding results in multiple mechanisms of action including increased antibody binding density, potent effector function, improved receptor internalization and HER2 downregulation relative to trastuzumab[4].

We present recent data that further explores zanidatamab's mechanistic differentiation compared to T, P and T + P.

Zanidatamab Binding Forms Unique HER2 Caps & Increased HER2 Cluster Size Compared to T + P on the Cell Surface

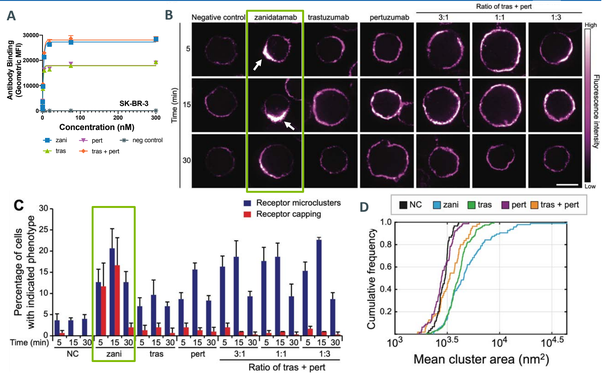
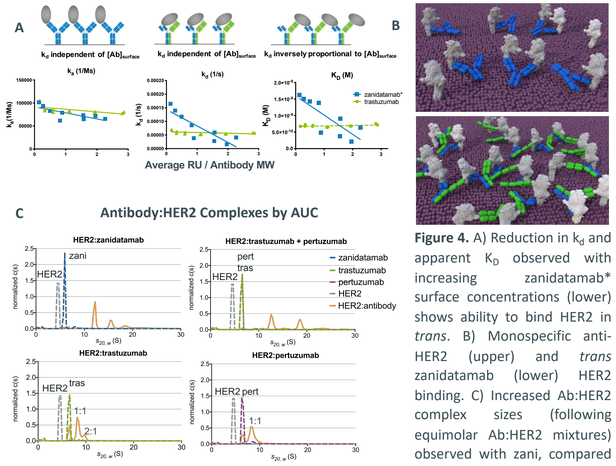


Figure 3. A) Zanidatamab and tras + pert (1:1) bind with increased Ab density on HER2-overexpressing SK-BR-3 by flow cytometry. B & C) Zanidatamab binding results in unique HER2 capping at 5 and 15 min (arrow) by confocal microscopy. D) dSTORM analysis shows increased HER2 cluster size following zani binding (15 min) than T, P, and T + P. B, C, D) HER2 localization detected on SK-BR-3 surface with anti-HER2-ECD1-AF647 Ab. See **Abstract #1032** for details.

Zanidatamab Binds HER2 in *trans* and Forms Large Antibody:HER2 Complexes in Solution



zanidatamab* analogue, (with non-affinity matured anti-ECD2) with equivalent apparent K_d compared to trastuzumab at low antibody capture, used to control for affinity/apparent affinity differences between zanidatamab and trastuzumab. Surface plasmon resonance (SPR), Response Units (RU), Molecular Weight (MW), Analytical Ultracentrifugation (AUC).

Figure 4. A) Reduction in k_d and apparent K_D observed with increasing zanidatamab* surface concentrations (lower) shows ability to bind HER2 in *trans*. B) Monospecific anti-HER2 (upper) and *trans* zanidatamab (lower) HER2 binding. C) Increased Ab:HER2 complex sizes (following equimolar Ab:HER2 mixtures) observed with zani, compared to tras or pert, shows ability to bind HER2 in *trans*.

Zanidatamab Elicits Superior CDC and Growth Inhibition Compared to Trastuzumab + Pertuzumab

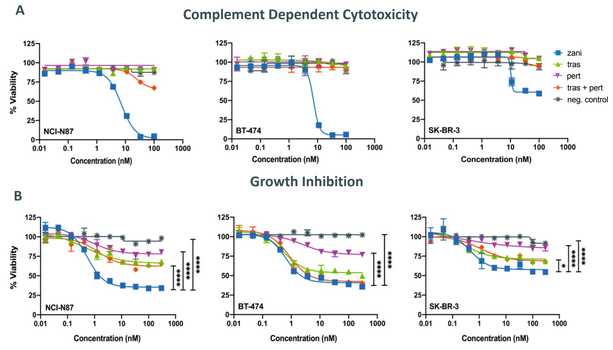


Figure 5. A) Zanidatamab mediates potent CDC with human complement serum in HER2-overexpressing tumor cells; trastuzumab, pertuzumab, and trastuzumab + pertuzumab (1:1) are inactive. B) Zanidatamab mediates potent tumor growth inhibition in HER2-overexpressing tumor cells and is superior to trastuzumab + pertuzumab (1:1) in NCI-N87 and SK-BR-3.

Zanidatamab Mediates Internalization, Receptor Depletion, ADCC and ADCP

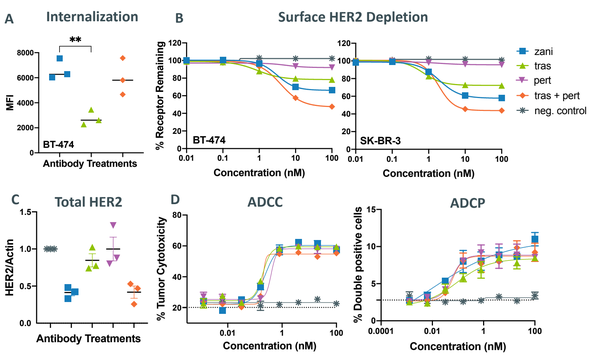
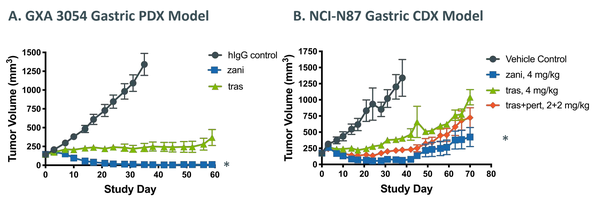


Figure 6. Zanidatamab treatment results in A) increased antibody internalization relative to tras (24 h), intracellular antibody detected by flow cytometry, B) surface HER2 depletion (24 h), surface HER2 detected with non-competing anti-HER2-ECD1-AF647 antibody following antibody treatment by flow cytometry, C) reduction of total HER2 (24 h) in SK-BR-3, determined by western immunoblotting, and D) potent ADCC and ADCP in SK-BR-3, using donor PBMC and donor-derived differentiated macrophage, respectively (dashed line represents no treatment control).

Zanidatamab has Superior Anti-Tumor Activity Compared to Anti-HER2 mAbs in HER2 3+ Gastric Cancer Models



- Total IgG serum exposure was similar for all test articles in each study (data not shown)

Xenograft Model HER2 Expression

Model	IHC HER2	FISH HER2
GXA-3054	3+	nd
NCI-N87	3+	Amplified

nd = not determined

Figure 7. A) All test articles administered at 30 mg/kg twice weekly for 5 weeks; B) Test articles administered at indicated equimolar dose levels twice weekly for four weeks. *, p<0.05 vs. trastuzumab & tras + pert by linear mixed-effects model fit to log-transformed tumor volumes over time. Differences in tumor growth rate between treatment groups was assessed using a Wald test.

Zanidatamab is a Promising Bispecific Antibody for the Treatment of HER2-Expressing Cancers

- Zanidatamab binds HER2 in *trans* and has multiple mechanisms of action that may collectively contribute to its anti-tumor activity
- Zanidatamab mediates unique HER2 capping and superior CDC and growth inhibition of HER2-overexpressing tumors compared to trastuzumab, pertuzumab, and trastuzumab + pertuzumab
- Zanidatamab is superior to trastuzumab and a combination of trastuzumab + pertuzumab in HER2-overexpressing gastric cancer xenografts
- Zanidatamab is actively being evaluated in clinical trials in multiple HER2 overexpressing solid tumors, including in HERIZON-BTC-01, a registration-enabling clinical trial in HER2 gene amplified biliary tract cancer (NCT04466891)

References

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