

# ZW220, a novel NaPi2b-targeting antibody drug conjugate bearing a topoisomerase I inhibitor payload

Andrea Hernández Rojas<sup>1</sup>, Jodi Wong<sup>1</sup>, Dunja Urošev<sup>1</sup>, Sam Lawn<sup>1</sup>, Kaylee J. Wu<sup>1</sup>, Saki Konomura<sup>1</sup>, Manuel Lasalle<sup>1</sup>, Diego A. Alonzo<sup>1</sup>, Luying Yang<sup>1</sup>, Mark Petersen<sup>1</sup>, Lemlem T. Degefiel<sup>1</sup>, Araba P. Sagoe-Wagner<sup>1</sup>, Sara Hersberger<sup>2</sup>, Sohyeong Kang<sup>1</sup>, Chi Wing Cheng<sup>1</sup>, Kevin Yin<sup>1</sup>, Raffaele Colombo<sup>1</sup>, Daya Siddappa<sup>1</sup>, Stuart D. Barnscher<sup>1</sup>, Jamie R. Rich<sup>1</sup>  
 Author affiliations: <sup>1</sup>Zymeworks Inc., Vancouver, BC, Canada; <sup>2</sup>ToxStrategies, LLC, Katy, TX, United States



## Introduction

### ADC structure

ZW220 is an antibody-drug conjugate (ADC) targeting human NaPi2b. The ADC is comprised of a novel fully humanized IgG1 antibody covalently conjugated to a novel topoisomerase I inhibitor payload, a camptothecin derivative, via endogenous interchain cysteines. The drug-linker consists of a maleimidocaproyl (MC) anchor and a GGFG-aminomethyl (AM) protease cleavable sequence. Drug to antibody ratios (DAR) of 8 and 4 have been evaluated with ZW220 ADC.

### Mechanism of action

Following NaPi2b binding and receptor-mediated internalization of ZW220, intracellular payload release induces targeted cell death in NaPi2b-positive cells, and subsequent death of NaPi2b-negative cells through bystander-mediated killing.

### ZW220 ADC

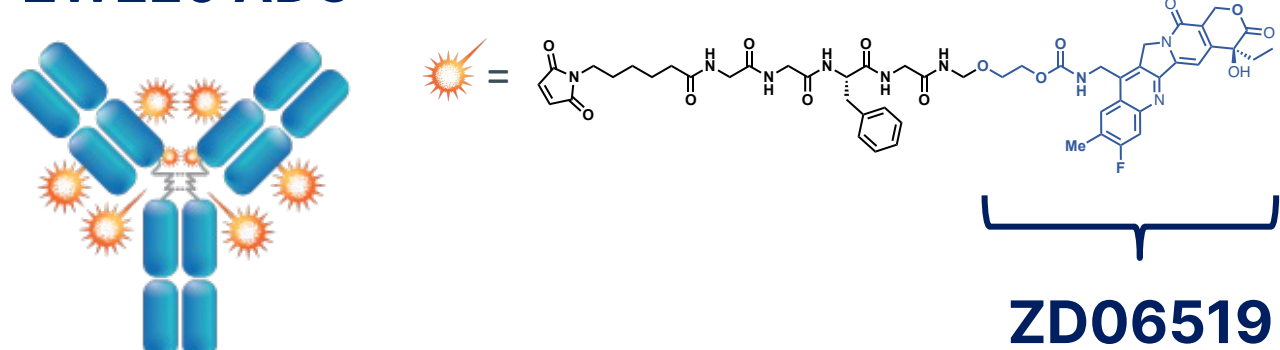


Figure 1. Structure of ZW220, a NaPi2b-targeting ADC comprised of novel monospecific IgG1 mAb, covalently conjugated to 8 or 4 drugs per antibody (MC-GGFG-AM-ZD06519 drug linker); DAR 8 pictured.

## NaPi2b is overexpressed in ovarian and lung cancers

### Expression

NaPi2b is highly expressed in ovarian and lung carcinomas; some NaPi2b expression is also found in endometrioid<sup>1</sup>, thyroid<sup>1</sup>, colorectal<sup>2</sup> and breast carcinomas<sup>3</sup>. Normal tissue expression of NaPi2b is observed in lung, liver, and small intestine.

### Function

NaPi2b is a multi-pass transmembrane sodium-dependent phosphate transport protein, encoded by SLC34A2 gene, involved in phosphate homeostasis.

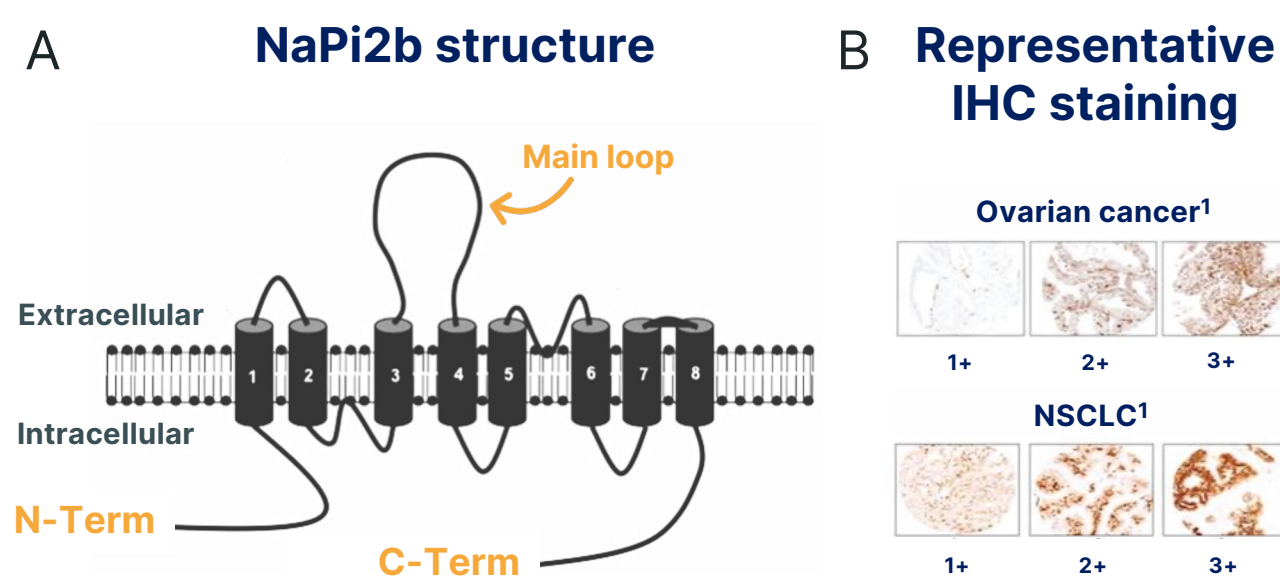
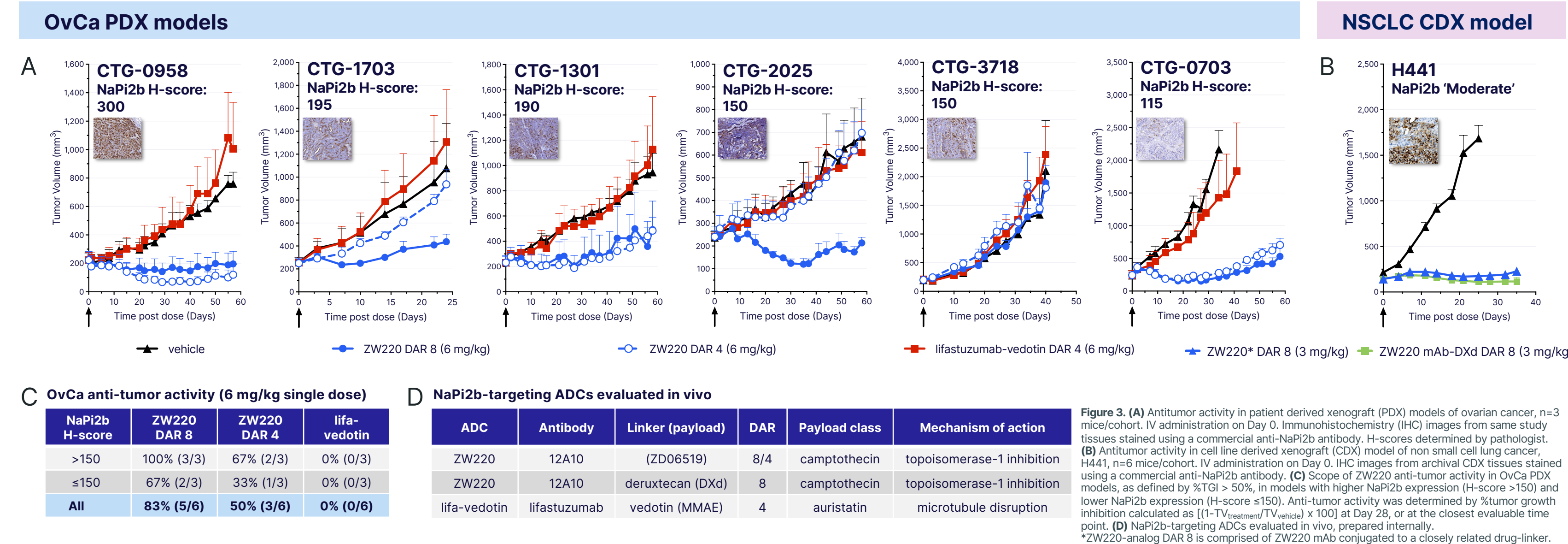


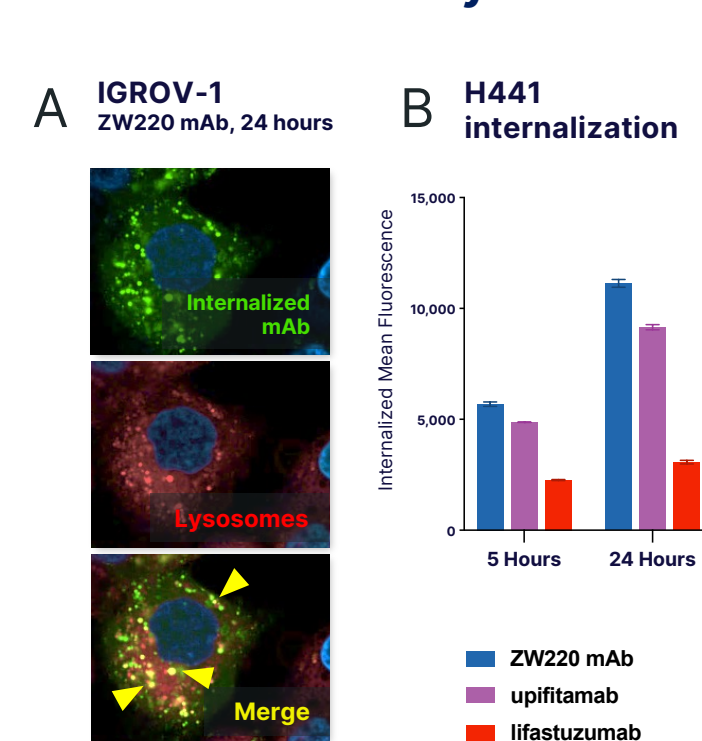
Figure 2. (A) Structural model of NaPi2b multi-pass transmembrane protein, adapted from Bobeck et al. 2015<sup>4</sup>. (B) Immunohistochemistry of representative tumor samples (stained using mouse NaPi2b antibodies) shows NaPi2b expression in human non-small cell lung cancer (NSCLC) and nonmucinous ovarian cancer.

## ZW220 demonstrates robust anti-tumor activity in ovarian carcinoma and NSCLC xenograft models with a range of NaPi2b-expression

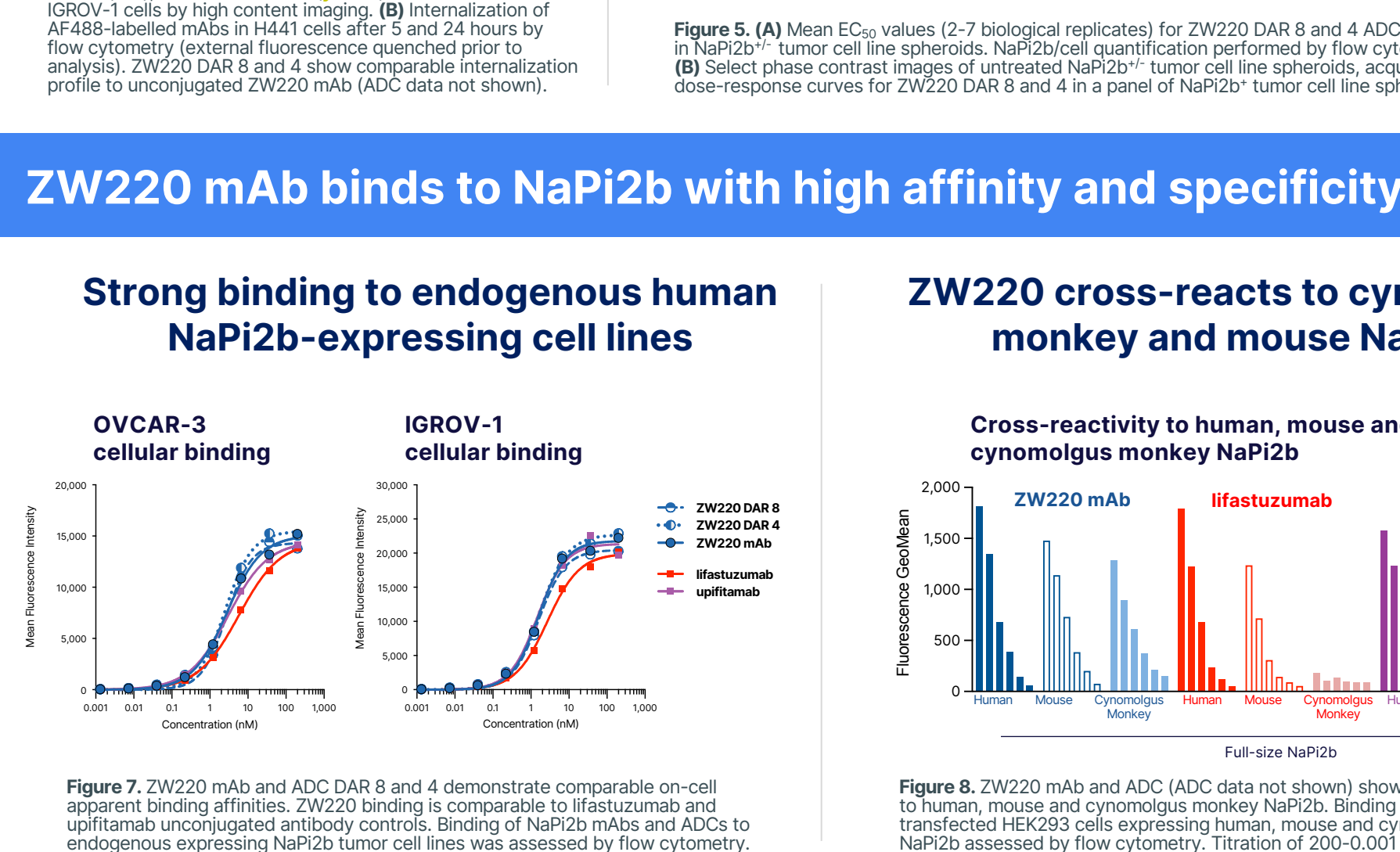


## ZW220 exhibits rapid internalization, potent target-mediated cytotoxicity and bystander killing in tumor cell lines

### ZW220 is efficiently internalized and colocalizes with lysosomes



## ZW220 mAb binds to NaPi2b with high affinity and specificity



### ZW220 induces cell growth inhibition in lung and ovarian NaPi2b+ tumor cell line spheroids

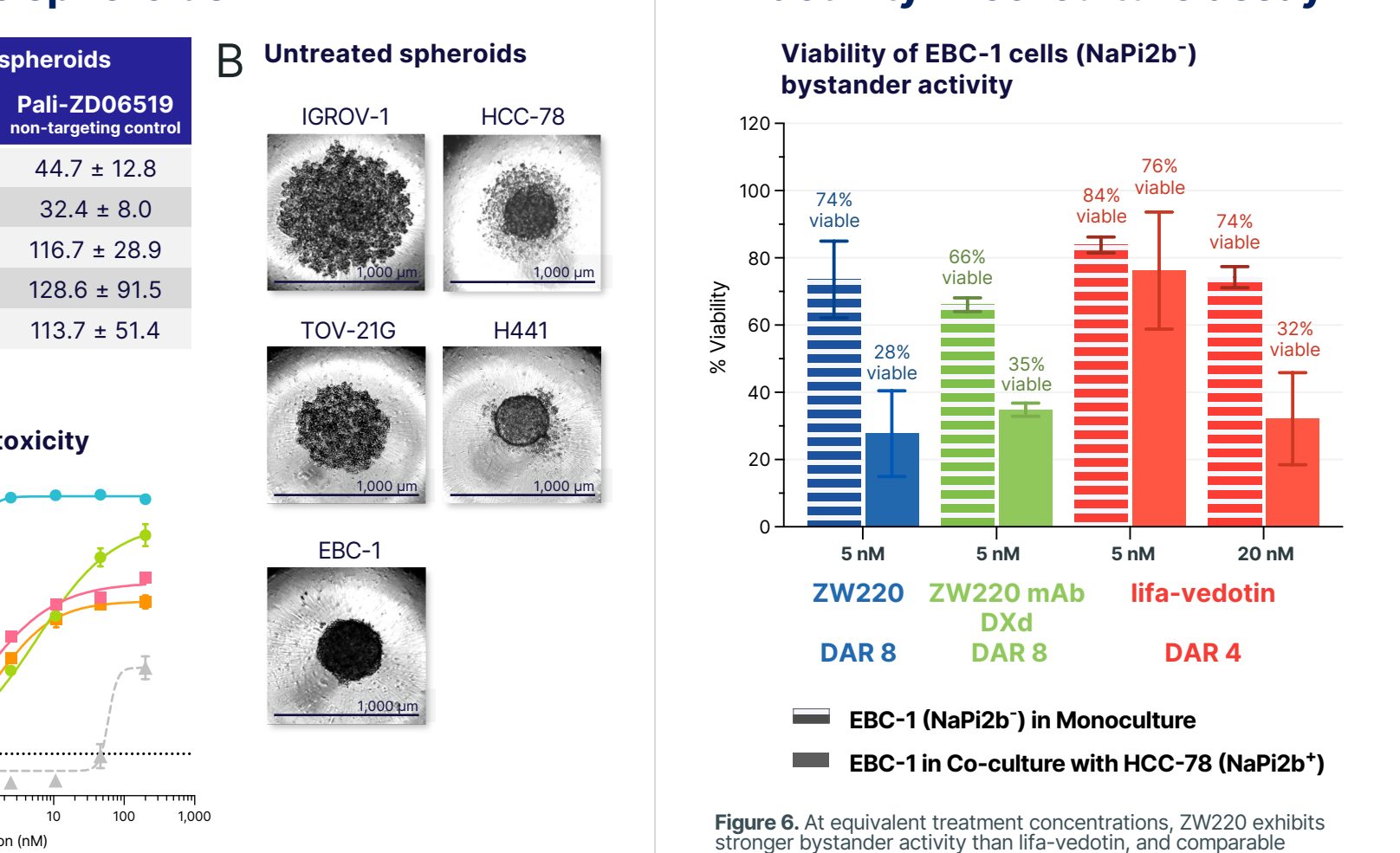
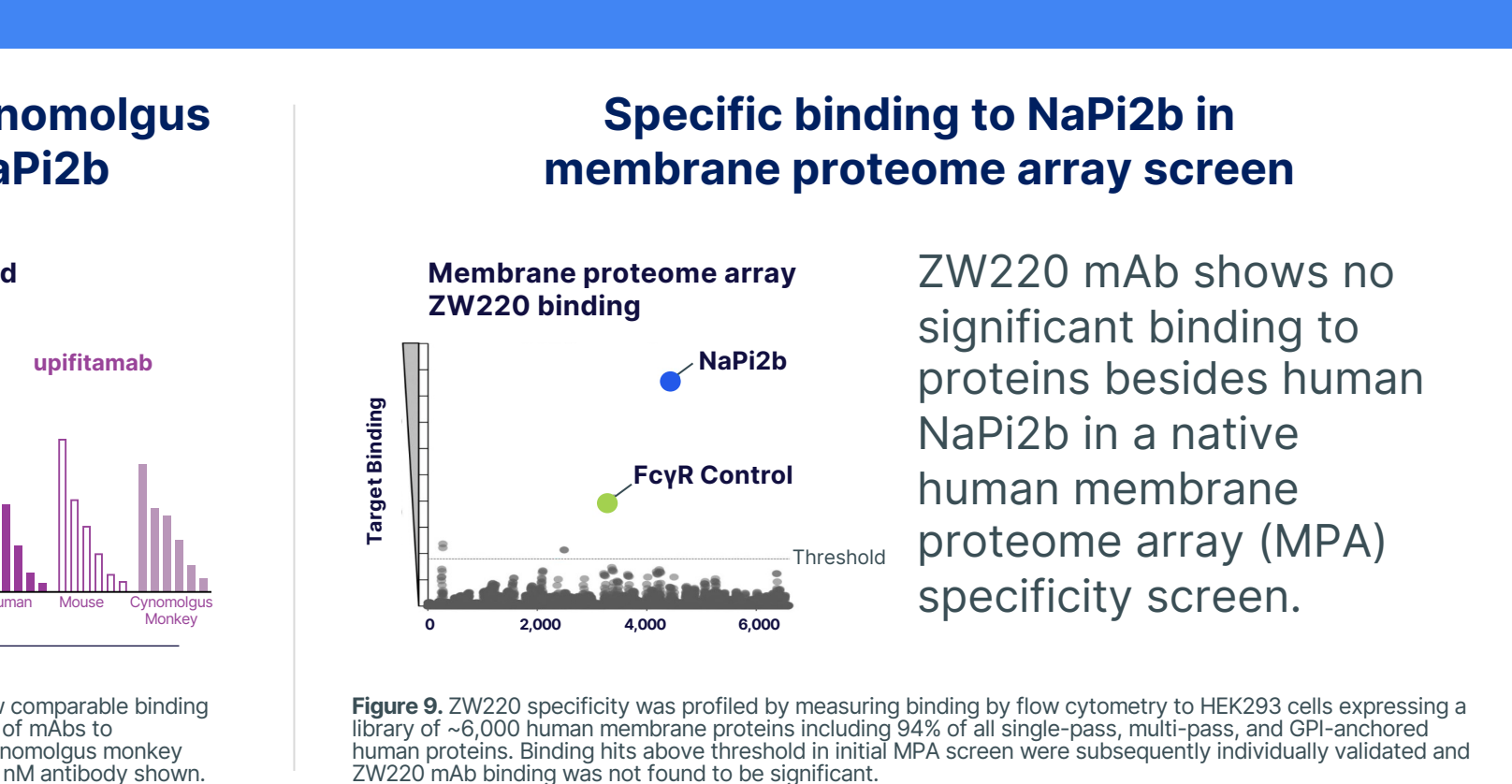


Figure 5. (A) Mean EC<sub>50</sub> values (2-7 biological replicates) for ZW220 DAR 8 and 4 ADCs and non-targeting control ADC. Pali-ZD06519 DAR 8, in NaPi2b<sup>-</sup> tumor cell line spheroids. NaPi2b/cell quantification performed by flow cytometry using AF47-labelled NaPi2b monoclonal antibody. (B) Select phase contrast images of untreated NaPi2b<sup>+</sup> tumor cell line spheroids, acquired moments prior to ADC treatment. (C) Representative dose-response curves for ZW220 DAR 8 and 4 in a panel of NaPi2b<sup>+</sup> tumor cell line spheroids and NaPi2b<sup>-</sup> tumor cell line spheroid control.

### ZW220 cross-reacts to cynomolgus monkey and mouse NaPi2b



## ZW220 is well tolerated in non-human primates

- Repeat-dose non-GLP toxicology study of ZW220 DAR 8 and DAR 4 in male NHPs resulted in no mortalities.
- No adverse clinical observations, effect on body weights, macroscopic observations or organ weights were noted.
- No clinical or anatomic pathology findings related to administration of ZW220.
- MTD of DAR 8 is 45 mg/kg; MTD of DAR 4 is 90 mg/kg.

Test article	Dose	Mortality	Clinical observations	Histo-pathology	Clinical chemistry	Hematology	MTD	T <sub>1/2</sub> (day)
ZW220 DAR 8	15 mg/kg	None	None	None	None	None	45 mg/kg	8.7
	30 mg/kg	None	None	None	None	None	45 mg/kg	7.7
	45 mg/kg	None	Fecal abnormalities (soft/loose)	None	None	None	45 mg/kg	8.0
ZW220 DAR 4	30 mg/kg	None	None	None	None	None	90 mg/kg	10.3
	60 mg/kg	None	None	None	None	None	90 mg/kg	9.8
	90 mg/kg	None	Fecal abnormalities (soft/loose/watery)	None	None	None	90 mg/kg	8.0

Figure 10. Relevant study parameters from a repeat dose non-GLP toxicology study in male cynomolgus monkeys performed to assess the tolerability and pharmacokinetic profile of ZW220 DAR 8 and 4 (n=3 animals/group). \*Half life (T<sub>1/2</sub>) calculated from Total IgG data in Figure 11B.

## ZW220 has a favorable pharmacokinetic profile

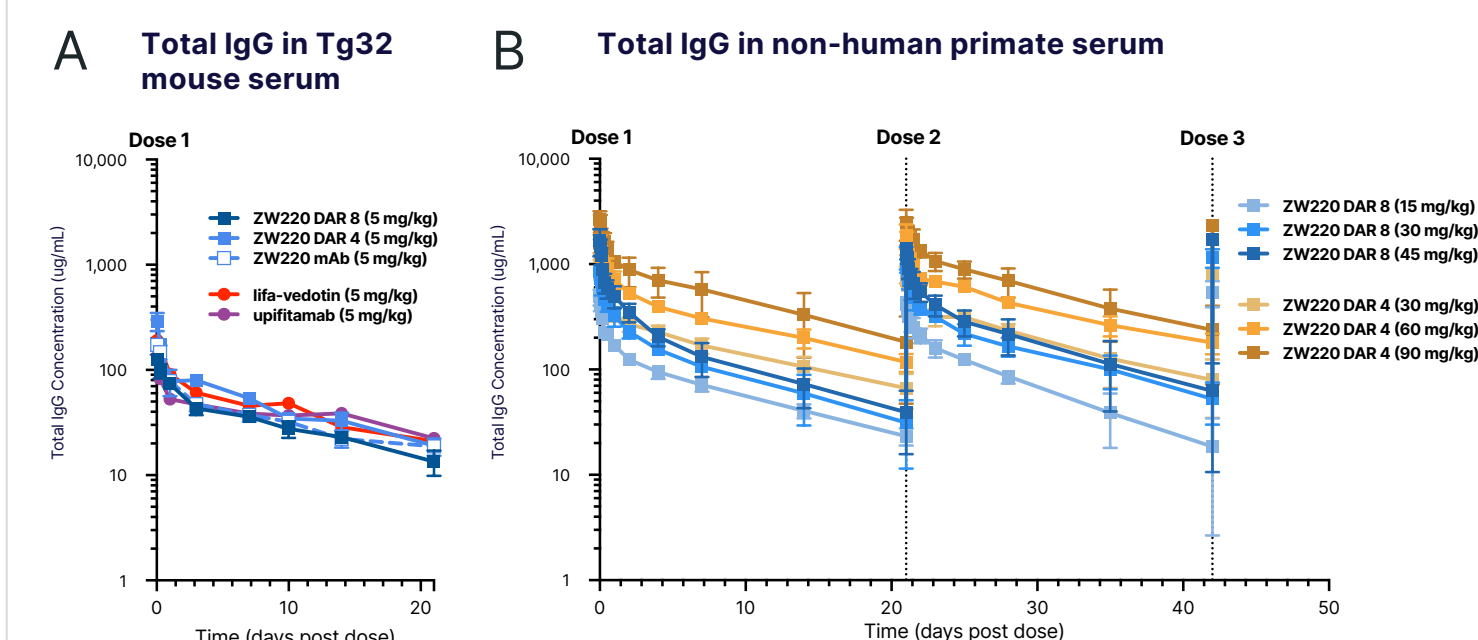


Figure 11. (A) Circulating antibody levels in Tg32 transgenic mouse model (expressing human neonatal Fc receptor, hFcRn) determined by ligand binding assay (LBD) measuring human IgG in serum following single intravenous dosing of antibody or ADC. (B) Circulating antibody levels in NHP determined by MSD assay following 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> dose (1<sup>st</sup> time point only).

## Conclusions

- ZW220 demonstrates robust preclinical anti-tumor activity in ovarian and lung cancer xenograft models with low NaPi2b expression levels (H-score ≥ 115).
- ZW220 is tolerated at high doses in non-human primates, with an MTD of 45 mg/kg for DAR 8 ADC, and 90 mg/kg for DAR 4 ADC.
- Potential for improvement over NaPi2b-targeting microtubule inhibitor-based ADCs on basis of efficacy, tolerability, and payload mechanism.
- Robust preclinical data package supports the continued development of ZW220 as a best-in-class NaPi2b ADC.

## References

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