Osimertinib treatment drives expression of TROP2, and combination treatment with datopotamab deruxtecan (Dato-DXd), a TROP2-directed antibody-drug conjugate, enhances its efficacy in PDX models of EGFR mutant non small-cell lung cancer

Matthew Martin[1]*, Alex Koers[1], Fernando Calero[1], Sara Talbot[1], Adina Hughes[1], Lukasz Magiera[1], Lucy Ireland[1], Benjamin Phillips[2], Ricardo Miragaia[3], Anca Farcas[1], Pablo Moreno[3], Ultan McDermott[1] and Nicolas Floc'h[1].

AstraZeneca – 1 Francis Crick Avenue; Cambridge, UK CB2 0AA. 1. Oncology Targeted Discovery, Bioscience 2. Data Sciences & Quantitative Biology, Discovery Sciences 3. Oncology Data Science *presenting author (*matthew.j.martin@astrazeneca.com*)

LB458

Introduction

- Osimertinib is a 3rd generation EGFR tyrosine kinase inhibitor with proven efficacy in the first- and second-line advanced or metastatic EGFR-mutant (EGFRm) NSCLC setting. Despite clinical benefit, most patients develop resistance to treatment, highlighting the need for combination strategies in both the front-line and post-progression settings, to maximize duration of response.
- Combining osimertinib with platinum-doublet chemotherapy has shown significant clinical benefit[1], prompting investigations into targeted delivery of chemotherapy via antibody-drug conjugates (ADCs).
- Datopotamab deruxtecan (Dato-DXd), a TROP2-directed ADC[2], has shown promising clinical activity as monotherapy EGFRm segment[3], thus we wished to understand how osimertinib treatment affects TROP2 expression.

Results

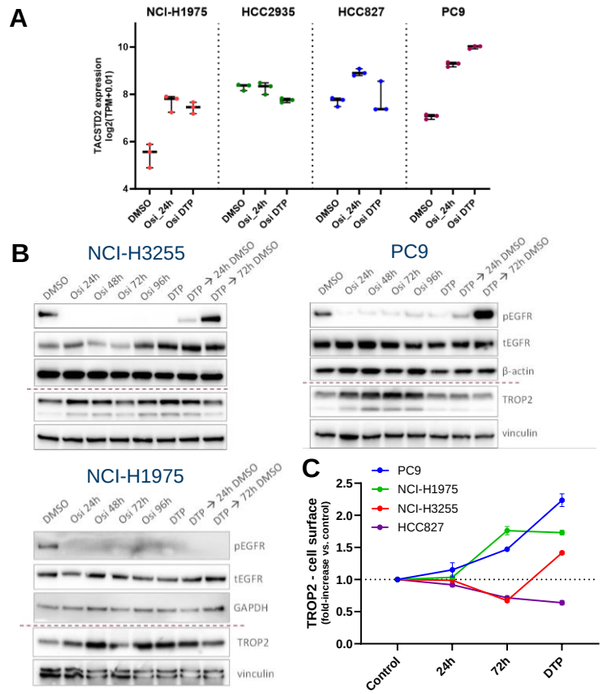

Fig. 1 Osimertinib treatment promotes upregulation of TROP2 at the mRNA and protein level. **A** Expression of the mRNA for TROP2 (*TACSTD2)* in 4 EGFRm cell lines treated with osimertinib for 24h (ACUTE) or 21 days (DTP)[4], plotted as log2 of transcripts per million (TPM). Expression of TROP2 protein, in whole cell lysates (**B**; western blot) or cell surface (**C**; flow cytometry) in EGFRm cell lines treated with osimertinib the indicated timeframes (DTP = 14 days treatment). Where indicated DTPs were released from drug for 24 or 72h (DMSO).

Results (ctd.)

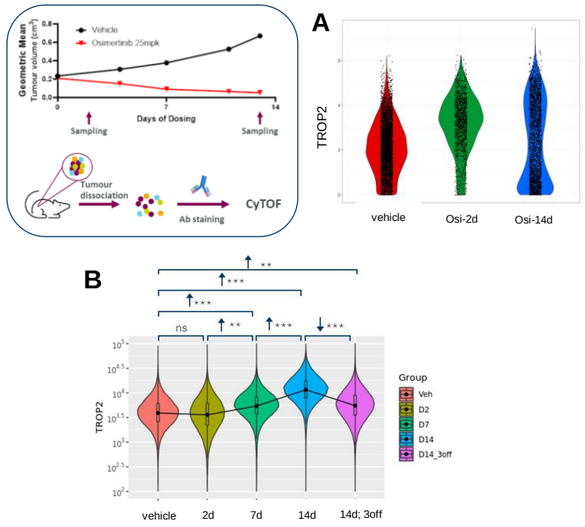

Fig. 2 TROP2 protein levels are increased in vivo upon osimertinib treatment **A** Expression of TROP2 in PC9 xenografts treated with osimertinib (25 mpk) for 2d or 14 days (DTP), measured by cytometer time of flight (CyTOF) mass spectrometry. **B** Measurement of cell surface TROP2 by flow cytometry in PC9 xenografts treated with osimertinib for 2, 7 and 14 days, as well as 14 days followed by 3 days off drug. ns = not significant; **p-adj <0.005 ***p-adj <0.001.

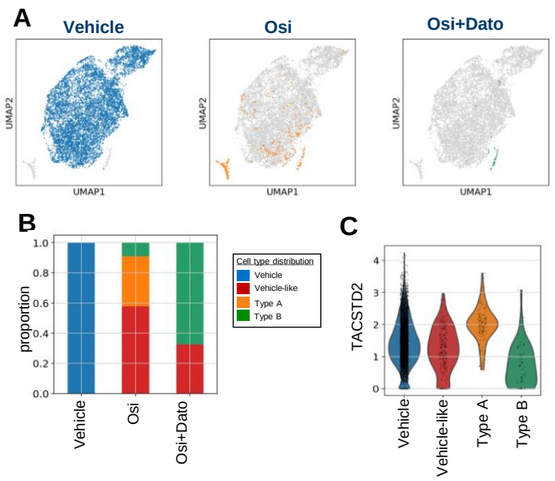

Fig. 3 The addition of Dato-DXd selectively eliminates the population of osimertinib persister cells with high TROP2 expression **A** UMAP clustering visualization of scRNAseq data from PC9 xenografts treated with osimertinib monotherapy, osimertinib + Dato-DXd (Dato) or vehicle control for 14days. **B** Cell type distribution of 3 treatment groups based on RNA expression across all samples. **C** Expression of *TACSTD2* in the indicated cell type.

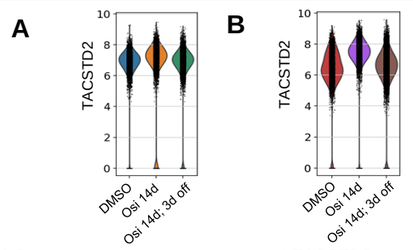

Fig. 4 Osimertinib induces upregulation of *TACSTD2* expression in EGFRm patient-derived organoids that is reversed upon drug removal. Expression of *TACSTD2* mRNA in scRNAseq datasets derived from *EGFR-L858R* patient derived organoids (**A**: HUB-07B2051; **B**: TEMPUS AZ574812) treated with vehicle (DMSO) vs. osimertinib for 14 days, with or without an additional 3 days in drug-free media.

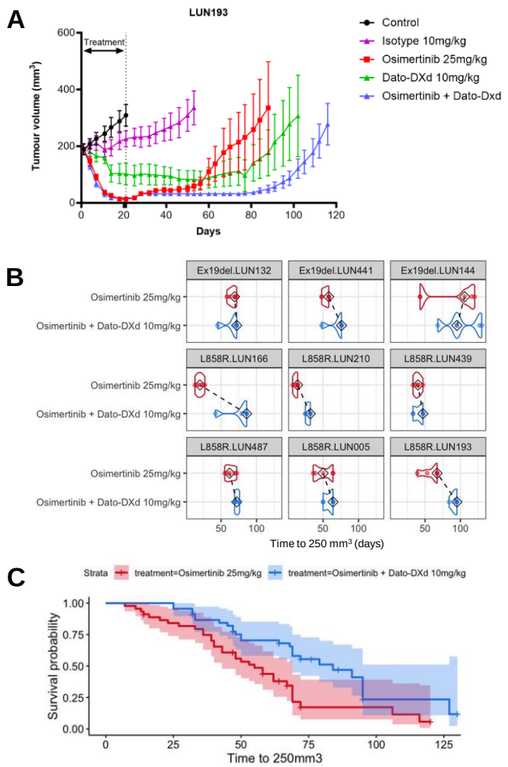

Fig. 5 Combining Osimertinib and Dato-DXd gives improved efficacy over Osimertinib monotherapy in a subset of EGFRm PDX models. **A** *In vivo* efficacy with the indicated treatments in the LUN193 PDX model. Antibody therapy was administered on Day 1, while osimertinib treatment continued daily for 21 days, after which there was a drug-free regrowth phase. **B** Graphical representation of the time to tumours reaching a size of 250 mm³, in days from the initiation of the experiment, across a panel of *EGFR*m PDX models treated with osimertinib monotherapy or the indicated Dato-DXd combination. **C** Survival probability, as determined by time to tumours reaching 250 mm³, across 9 EGFRm PDX models for the indicated treatment groups.

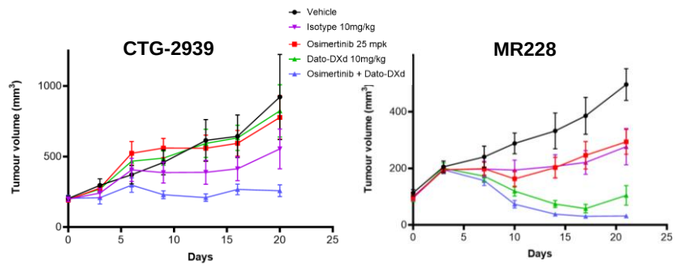

Fig. 6 Combining osimertinib and Dato-DXd shows improved efficacy over either drug alone in Osimertinib-resistant PDX models. Tumour-bearing animals were treated as indicated with osimertinib, Dato-DXd, vehicle or antibody isotype control. Antibody therapy was administered on Day 1, while osimertinib was given daily throughout the experiment.

Table 1 Tumour growth inhibition (TGI %) of osimertinib/Dato-DXd combination in PDX models from progressed patients

Model	Osi	Isotype	Dato-Dxd	Osi + Dato-Dxd
CTG-2939	1%	46%	0%	92%
DFCI-403	0%	26%	32%	39%
MR228	42%	50%	105%	167%
MR260	155%	123%	150%	182%

Conclusions

- Osimertinib treatment leads to increased expression of TROP2 at the mRNA and protein level, translating to enhanced levels at the cell surface.
- Increased TROP2 expression is reversed upon drug withdrawal.
- scRNAseq of PC9 xenografts shows a subset of osimertinib residual cells have high TACSTD2 expression, and this cell population is eliminated by co-treatment with Dato-DXd.
- The Dato-DXd/osimertinib combination leads to improved efficacy over osimertinib monotherapy in 4/9 first-line EGFRm PDX models tested.
- Combination therapy showed benefit over either agent alone in 3/4 PDX models derived from patients who relapsed on osimertinib.
- Together these data support ongoing clinical testing of the osimertinib-Dato-DXd combination in the first-line (TL-14) and progression (TL-15) settings.

References

1. Planchard et al., *N Engl J Med* ***389***(21): 1935-1948, 2023.
2. Okajima et al., *Mol Cancer Ther* **20**(12): 2329-40, 2021.
3. TROPION-LUNG-01 and TROPION-LUNG-05.
4. Criscione et al., *NPJ Precision Oncology* **6**(1): 95-107, 2022.

Acknowledgements

This study was sponsored by AstraZeneca. In July 2020, Daiichi-Sankyo entered into a global development and commercialisation collaboration with AstraZeneca for datopotamab deruxtecan (Dato-DXd).