Poster 5928

Baseline serum protein analysis of patients with interstitial lung disease/pneumonitis (ILD) in 3 trastuzumab deruxtecan (T-DXd) trials: DESTINY-Breast01, DESTINY-Breast04, and DESTINY-Lung01

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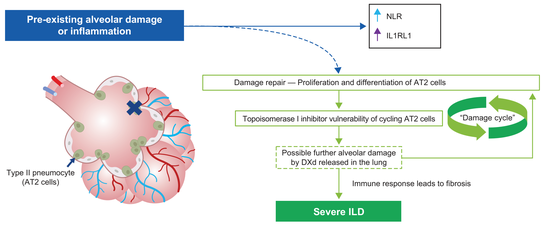
Objective

- This exploratory analysis aimed to identify candidate interstitial lung disease/pneumonitis (ILD) baseline risk biomarkers via proteomic analysis and evaluate Krebs von den Lungen-6 (KL-6) levels in serum samples from patients who received T-DXd treatment in the DESTINY-Breast01 (DB-01), DESTINY-Breast04 (DB-04), and DESTINY-Lung01 (DL-01) trials

Conclusions

- Proteomics analysis identified elevated baseline serum interleukin 1 receptor-like 1 (IL1RL1; also known as ST2) levels as a potential baseline marker for T-DXd–related grade 3-5 ILD in patients with lung or breast cancer
- A trend towards elevated baseline IL1RL1 was observed in patients with oxygen saturation (SpO_2) <95%; additionally, a correlation between elevated IL1RL1 and the neutrophil-lymphocyte ratio (NLR) was observed, suggesting a possible link between elevated baseline IL1RL1 and compromised lung function, as well as the presence of inflammation (**Figure 1**)
- Although KL-6 has been previously reported as an ILD biomarker,[1-4] our analysis did not demonstrate a consistent elevation of KL-6 in ILD patients across the 3 studies evaluated, potentially due to the baseline elevation of KL-6 in these patient populations irrespective of ILD status
- Given the limited number of clinical studies and the few grade 3-5 ILD events included in this exploratory analysis, results should be interpreted with caution. Further investigation is warranted to explore the utility of baseline IL1RL1 levels as a predictive biomarker for T-DXd-related severe ILD risk

Figure 1. Results driven hypothesis: Elevation of IL1RL1 may predict ILD occurrence



Plain Language Summary

Why did we perform this research?
ILD is a complex group of lung disorders that can develop as an adverse event in patients receiving T-DXd cancer therapy. The exact cause of ILD in these patients remains unknown, and the underlying mechanisms triggering its development are not well understood. Gaining insights into these mechanisms and finding ways to diagnose ILD earlier and better could lead to improved practices of ILD prevention and treatment.

How did we perform this research?
We analyzed specific protein levels in the blood of patients before T-DXd treatment and compared the results between those patients who later developed ILD and those patients who did not. The goal was to identify potential markers for ILD predisposition and to better understand the underlying biological pathways.

What were the findings of this research?
This study found that higher pretreatment levels of the protein IL1RL1 may be indicative of severe ILD events in patients receiving T-DXd. Patients with blood oxygen levels below 95% tended to have higher IL1RL1 levels compared with those with blood oxygen levels above 95%. KL-6 has previously been considered as a biomarker for ILD, but our analysis did not find consistent elevations in KL-6 levels among patients treated with T-DXd with ILD across the 3 studies examined.

What are the implications of this research?
High IL1RL1 protein levels may help identify patients predisposed to developing severe ILD even before initiating T-DXd treatment. This would enable better monitoring, possible prevention, early detection, and optimization of care and ILD management. Insights into IL1RL1 and KL-6 pathways support future research aimed at improving patient health outcomes.

Where can I access more information?
To learn more about the trials included in this study please visit:
DESTINY-Breast01: https://clinicaltrials.gov/study/NCT03248492.
DESTINY-Breast04: https://clinicaltrials.gov/study/NCT03734029.
DESTINY-Lung01: https://clinicaltrials.gov/study/NCT03505710.

Poster
Supplementary material

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This study was sponsored by Daiichi Sankyo. In March 2019, AstraZeneca entered into a global development and commercialization agreement with Daiichi Sankyo for trastuzumab deruxtecan (T-DXd; DS-8201).

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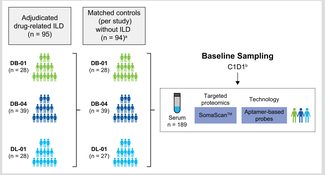
Introduction

- T-DXd is a human epidermal growth factor receptor 2 (HER2)-directed antibody-drug conjugate approved to treat patients with HER2-positive (HER2+), HER2-low, and HER2-ultra low metastatic breast cancer, HER2+ metastatic gastric cancer, *HER2*-mutant metastatic non–small cell lung cancer, and HER2+ (immunohistochemistry [IHC] 3+) metastatic solid tumors[5,6]
- ILD, a heterogeneous group of lung disorders that manifest as inflammation and/or fibrosis of the lungs, is an important known adverse event of special interest for T-DXd[7-9]
  - The precise mechanism of T-DXd–related ILD remains unknown
- Understanding the pathophysiology of drug-induced ILD and identifying biomarkers for patients at higher risk of T-DXd–related ILD is crucial for the safety and the efficient treatment of patients
- KL-6 has previously been analyzed as a biomarker for ILD,[1-4] but its relevance to T-DXd–related ILD has not been well studied and remains undetermined

Methods

Exploratory SomaLogic, SomaScan™ Analysis

Figure 2. SomaScan study design



[a]Baseline sample was missing for 1 control; therefore, only 94 matched controls were included.
[b]Baseline serum samples were collected before dosing in cycle 1.

- Baseline serum samples were collected from 189 patients in 3 T-DXd clinical trials (**Figure 2**):
  - **DB-01**: n = 56; NCT03248492 (T-DXd 5.4 mg/kg dose cohort)
  - **DB-04**: n = 78; NCT03734029 (T-DXd 5.4 mg/kg dose cohort)
  - **DL-01**: n = 55; NCT03505710 (T-DXd 5.4 mg/kg and 6.4 mg/kg dose cohorts)
- The selection criteria for studies included in this analysis were based on trial completion status, available data on reported ILD incidence, and patient consent for exploratory analysis
- All T-DXd–treated and consenting patients with adjudicated drug-related ILD (n = 95) were included and classified based on their worst adjudicated ILD grade
- T-DXd–treated matching controls, who did not develop ILD (n = 94), were then selected based on the following patient characteristics:
  - **DB-01:** best overall response (BOR), age, country, and study duration
  - **DB-04:** BOR, age, country, and sex
  - **DL-01:** BOR, age, country, sex, and dose cohort
- Available baseline serum samples were analyzed using the SomaLogic SomaScan™ 7k proteomics platform, employing up to 7289 SOMAmer (slow off-rate modified aptamer) single-stranded DNA reagents to profile unique human proteins
  - The assay used 12 hybridization normalization control sequences, 5 pooled human calibrator controls, and 3 pooled quality control (QC) replicates
    - Any sample with a SOMAmer QC ratio on any plate outside the accepted accuracy range of 0.8-1.2, when compared with the reference,[10] failed QC features and was excluded from the analysis
      - The QC ratio is calculated as the ratio of the QC reference value in relative fluorescence units (RFU) to the median RFU value of QC replicates for each SOMAmer on a given plate[10]
- Proteins associated with ILD severity were identified through differential expression analysis. Protein level distributions and unadjusted *P* values from the Kruskal-Wallis test were used to evaluate differences among ILD severity groups

Confirmatory Luminex™ Analysis

- Absolute quantification of serum IL1RL1 levels was performed using the Luminex™ platform with the custom Luminex® Discovery Assay Human Premixed Multi-Analyte Kit, comprising 9 magnetic bead assays (LXSAHM-08, R&D Systems, Inc.)
- Baseline serum samples were collected from a total of 608 patients in the 3 T-DXd clinical trials:
  - **DB-01:** n = 183; 29 patients with adjudicated drug-related ILD, 154 patients with no ILD
  - **DB-04:** n = 291; 39 patients with adjudicated drug-related ILD, 252 patients with no ILD
  - **DL-01:** n = 134; 28 patients with adjudicated drug-related ILD, 106 patients with no ILD

KL-6 Chemiluminescent Enzyme Immunoassay (CLEIA)

- KL-6 is an accepted biomarker for ILD in Japan,[11] often measured in the clinical setting. The normal range of serum KL-6 levels in healthy individuals is <500 U/mL[1]
- Given that KL-6 was a specific glycosylated form of MUC1 that was not included in SomaScan platform, serum KL-6 levels were measured by a proprietary CLEIA (SRL, Inc., Japan) at baseline before dosing in cycle 1 and longitudinally before dosing across cycles 2, 3, and 4, and then every 2 cycles until end of treatment
- Baseline and longitudinal serum samples were collected from 637 patients in the 3 T-DXd clinical trials:
  - **DB-01:** n = 182; 28 patients with adjudicated drug-related ILD, 154 patients with no ILD
  - **DB-04:** n = 321; 40 patients with adjudicated drug-related ILD, 281 patients with no ILD
  - **DL-01:** n = 134; 28 patients with adjudicated drug-related ILD, 106 patients with no ILD

Results

SomaScan Analysis: Patients by Worst Adjudicated ILD Grade

- Of the 95 patients with adjudicated drug-related ILD included in the SomaScan analysis, 74 had mild/moderate ILD (grades 1 or 2) and 21 had severe ILD (grades 3-5) (**Table 1**)

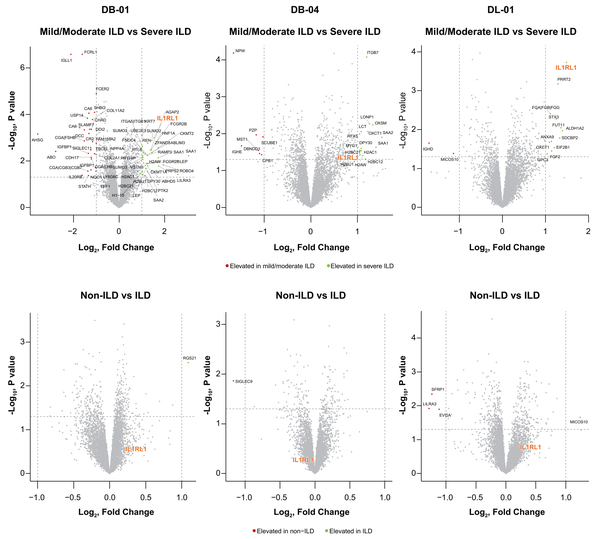
Table 1. Adjudicated drug-related ILD breakdown in patients included in the SomaScan proteomics analyses

n	DB-01 n = 56	DB-04 n = 78	DL-01 n = 55
Non-ILD	28	39	27
Adjudicated drug-related ILD	28	39	28
Grade 1	6	12	3
Grade 2	16	20	17
Grade 3	1	4	3
Grade 4	0	0	0
Grade 5	5	3	5

SomaScan: Differentially Expressed Proteins

- In total, 7289 unique analytes were evaluated, and 6204 analytes passed the SOMAmer QC across all 3 studies
  - In the DB-01 study, 7.2% (527/7289) of analytes failed QC
  - In the DB-04 study, 10.2% (740/7289) of analytes failed QC
  - In the DL-01 study, 6.5% (473/7289) of analytes failed QC
- From the 6204 analytes tested, elevated baseline serum IL1RL1 levels were observed in patients who later developed grade 3-5 ILD in all 3 studies (**Figure 3**)
  - When non-ILD cases were compared with all-grade ILD cases, no marker showed a consistently elevated pattern in these 3 studies (**Figure 3**)
  - Baseline IL1RL1 levels were significantly higher in patients who later developed grade 3-5 ILD compared with those who never developed ILD, based on data from a combined cohort of the 3 trials (data not shown)
- IL1RL1 was selected to be investigated further due to the higher elevated pattern observed in the SomaScan™ analysis in patients with severe ILD across the 3 studies

Figure 3. Differentially expressed proteins in patients from DB-01, DB-04, and DL-01

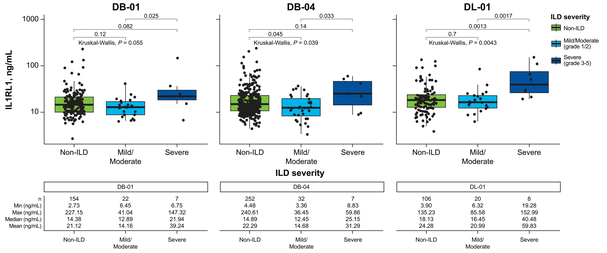


Highlighted proteins have *P* value <0.05 and absolute fold change >2.00

IL1RL1 Measurement by Luminex Assay

- Across the 3 studies, median levels of baseline IL1RL1 in non-ILD patients were comparable to grade 1/2 ILD cases; in contrast, the median baseline IL1RL1 level in grade 3-5 ILD cases was elevated (**Figure 4**)

Figure 4. IL1RL1 levels by ILD severity and study

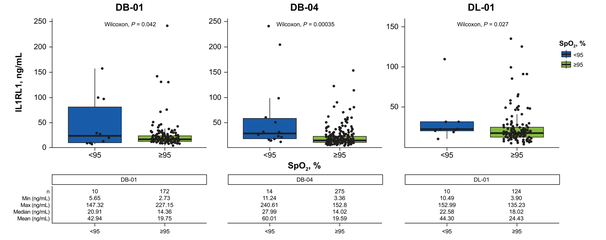


	DB-01			DB-04			DL-01		
n	154	22	7	252	32	7	106	20	8
Min (ng/mL)	2.73	6.45	6.75	4.48	3.36	8.83	3.90	6.32	19.28
Max (ng/mL)	227.15	41.04	147.32	240.61	36.45	59.86	135.23	85.58	152.99
Median (ng/mL)	14.38	12.89	21.94	14.89	12.45	25.15	18.13	16.45	40.48
Mean (ng/mL)	21.12	14.16	39.24	22.29	14.68	31.29	24.28	20.99	59.83

The number of patients with ILD included in each test varies based on the availability of patient samples for specific assays.

- There was elevated baseline IL1RL1 in patients with SpO_2 <95% (**Figure 5**)

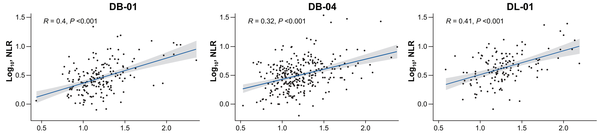
Figure 5. Baseline IL1RL1 level and SpO_2



	DB-01		DB-04		DL-01	
n	10	172	14	275	10	124
Min (ng/mL)	5.65	2.73	11.24	3.36	10.49	3.90
Max (ng/mL)	147.32	227.15	240.61	152.8	152.99	135.23
Median (ng/mL)	20.91	14.36	27.99	14.02	22.58	18.02
Mean (ng/mL)	42.94	19.75	60.01	19.59	44.30	24.43

- Baseline IL1RL1 level had a weak to moderate positive correlation with NLR. Both elevated IL1RL1 and NLR may reflect the presence of inflammation (**Figure 6**)

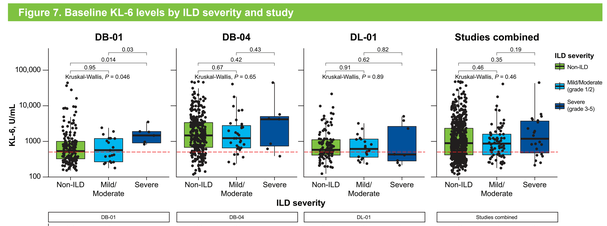
Figure 6. Baseline IL1RL1 level and NLR



Serum KL-6 by CLEIA

- The majority of patients had elevated baseline serum KL-6 levels (≥500 U/mL)[1] in all 3 studies:
  - **DB-01:** median, 527.5 U/mL; range, 154.0-44,400 U/mL
  - **DB-04:** median, 1470.0 U/mL; range, 119.0-48,100 U/mL
  - **DL-01:** median, 569.5 U/mL; range, 124.0-21,500 U/mL
- DB-01 patients with grade 3-5 ILD had statistically significant higher baseline KL-6 levels compared with those without ILD or with grade 1/2 ILD. This pattern was not consistently observed in the other 2 studies (**Figure 7**)
- Longitudinal sample analysis showed that KL-6 levels decreased with T-DXd treatment in selected DB-04 patients, mirroring the reduction in tumor burden observed during treatment (**Figure 8**)

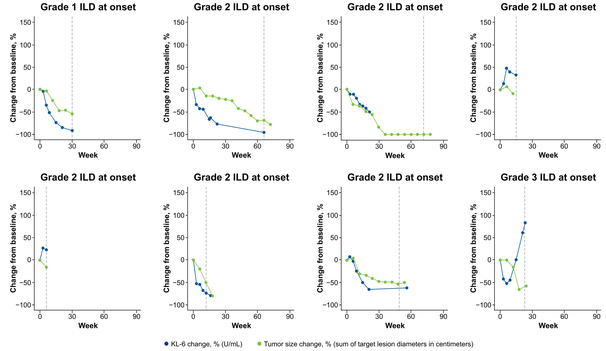
Figure 7. Baseline KL-6 levels by ILD severity and study



	DB-01			DB-04			DL-01			Studies combined		
n	154	21	7	281	33	7	106	20	8	541	74	22
Min (U/mL)	154	179	824	119	234	386	124	238	214	119	179	214
Max (U/mL)	44,400	2410	3490	48,100	41,100	44,700	21,500	3200	5050	48,100	41,100	44,700
Median (U/mL)	527.5	563.0	1460.0	1470.0	1220.0	4180.0	569.5	614.0	424.5	889.0	866.0	1212.5
Mean (U/mL)	1911.19	807.62	1617.57	3634.37	3083.21	8689.71	1390.11	880.15	1598.12	2735.29	2109.58	3960.73

The number of patients with ILD included in each test varies based on the availability of patient samples for specific assays. The dotted horizontal red line denotes the upper limit of the normal range of serum KL-6 levels in healthy individuals (<500.0 U/mL).[1]

Figure 8. Percentage change from baseline in KL-6 levels (U/mL) and tumor size (sum of target lesion diameters) in selected DB-04 patients with ILD and baseline KL-6 levels >75th percentile



The vertical dotted line denotes time of ILD onset. Each panel represents a different patient.

Abbreviations

AT2, alveolar type 2 progenitor cells; BOR, best overall response; C, cycle; CLEIA, chemiluminescent enzyme immunoassay; D, day; DB-01, DESTINY-Breast01; DB-04, DESTINY-Breast04; DL-01, DESTINY-Lung01; G, grade; HER2, human epidermal growth factor receptor 2; ILD, interstitial lung disease/pneumonitis; IL1RL1, interleukin 1 receptor-like 1; KL-6, Krebs von den Lungen-6; NA, not applicable; NLR, neutrophil-lymphocyte ratio; RFU, relative fluorescence unit; Q, quartile; QC, quality control; SpO_2, peripheral capillary oxygen saturation; SOMAmer, slow off-rate modified aptamer; T-DXd, trastuzumab deruxtecan.

Acknowledgments

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Disclosures

Z Tsuchihashi, K Kubota, J Zhao, X Wang, V Kumar, Y Cheng, T Kaji, and M Nakamura are employees of Daiichi Sankyo. NF Blasco and B Jakubison are employees of AstraZeneca. K. Contrepois, P. Newham, and S Terrillon are former employees of AstraZeneca. CA Powell discloses funding by AstraZeneca and consulting fees from Daiichi Sankyo, AstraZeneca, BioNTech, Duality, Pfizer, Seagen, and Merus. EF Smit discloses consulting fees from Daiichi Sankyo, AstraZeneca, Bristol Myers Squibb, Boehringer Ingelheim, Eli Lilly, Janssen, MSD, Roche, Sanofi, Takeda, and Merck. S Modi discloses funding and consulting fees from AstraZeneca, Daiichi Sankyo, Genentech, D3 Bio, Duality Bio, Nuvation, Pfizer, and Seagen, and consulting fees from Zymeworks and Avacta.

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