Case Study

MindWalk™

LensAI™ Epitope Mapping

LensAI Epitope Mapping matches x-ray crystallography

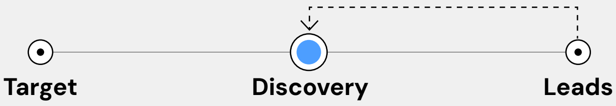
Outperforms other epitope mapping technologies in benchmark study

Overview:

Epitope mapping remains a cornerstone of therapeutic antibody development, and the 2023 peer-reviewed study "Epitope mapping of monoclonal antibodies" (published in MABS, 2023, Vol. 15, No. 1, 2285285) offers a rare head-to-head comparison of leading technologies across five high-impact antibody-antigen pairs. Evaluating seven experimental methods—from peptide arrays to hydrogen-deuterium exchange—the study highlights both the strengths and limitations of traditional approaches. Building on this benchmark, we applied LensAI *in silico* Epitope Mapping to go head-to-head and analyze the results.

Challenge:

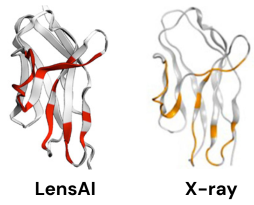
Traditional epitope mapping methods, such as x-ray crystallography and mass spectrometry, are time consuming, costly, and with feasibility that is highly dependent on the target type. Such challenges combined with the low-throughput nature of these technologies limit the utility of these methods to late-stage lead characterization or supporting IP filings. In contrast, *in silico* epitope mapping, which only requires sequences alone and is not reliant on physical material, offers a fast, high-throughput alternative that can be integrated earlier into the workflow, enabling more informed decisions and reducing risk. This case study examines the performance of LensAI *in silico* Epitope Mapping to demonstrate its potential for improving efficiency and confidence in the discovery process.

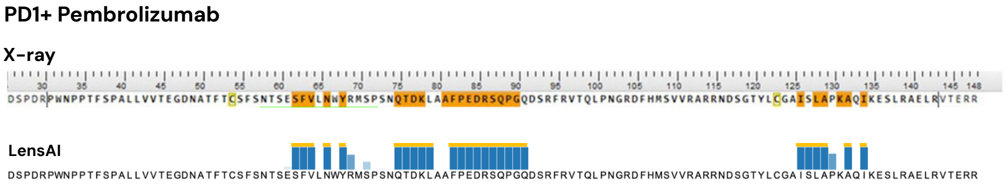


Background: a head-to-head comparison with x-ray crystallography and 6 additional technologies

The accuracy of LensAI Epitope Mapping is compared with x-ray crystallography, considered the gold standard, and six other methods (peptide array, alanine scan, domain exchange, hydrogen-deuterium exchange, chemical cross-linking, and hydroxyl radical footprinting) for epitope identification in five antibody-antigen combinations: Pembrolizumab+PD1, Nivolumab+PD1, Ipilimumab+CTLA4, Tremelimumab+CTLA4, and MK-5890+CD27.

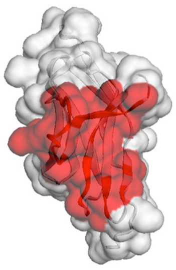
LensAI matched x-ray crystallography with exceptional accuracy

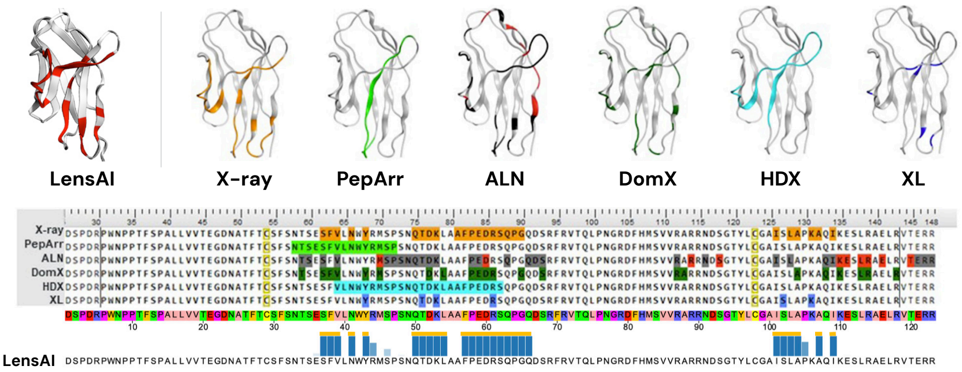




PD1 + Pembrolizumab

LensAI AUC = 0.80





AUC = 1 means a perfect prediction. AUC = 0.5 means no better than random guessing.

🟥 **3D target model:** intense red indicates epitope

🟦 **Blue bars:** height indicates the confidence score

🟨 **Orange line:** indicates hitting the epitope threshold

LensAI Epitope Mapping assigns a confidence score (0–1) to each amino acid (AA) in the target. Residues with scores above a set threshold (orange line) are classified as part of the predicted epitope. LensAI confidence scores are visualized as blue bars. The model's ability to distinguish epitopes from non-epitopes is measured by the AUC (Area Under the Curve), which plots the True Positive Rate against the False Positive Rate.

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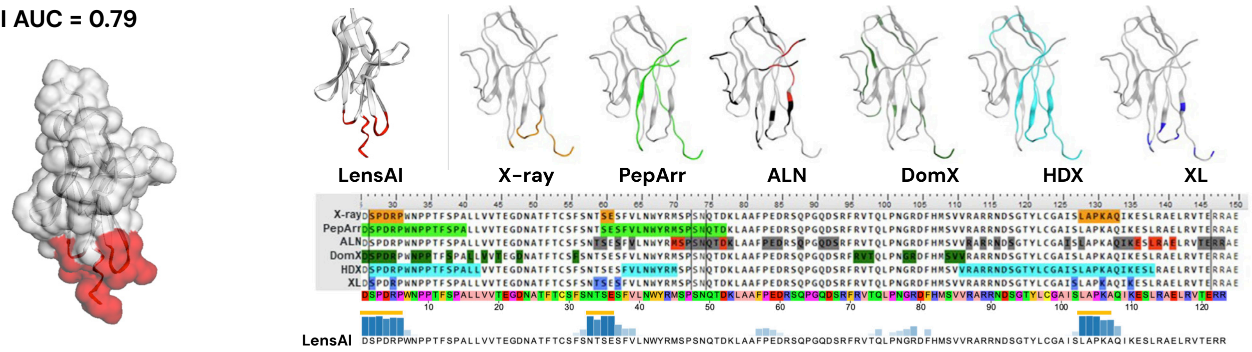
The benchmark comparison:

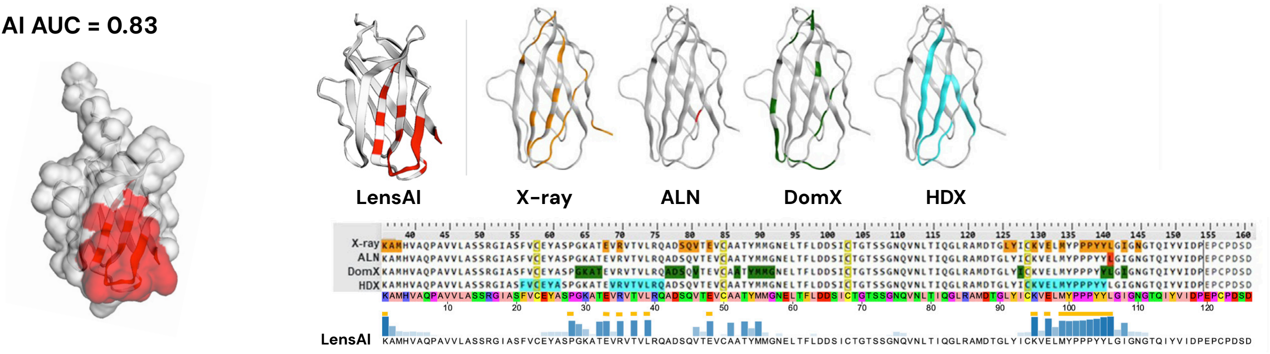
PD1 + Nivolumab

LensAI AUC = 0.79



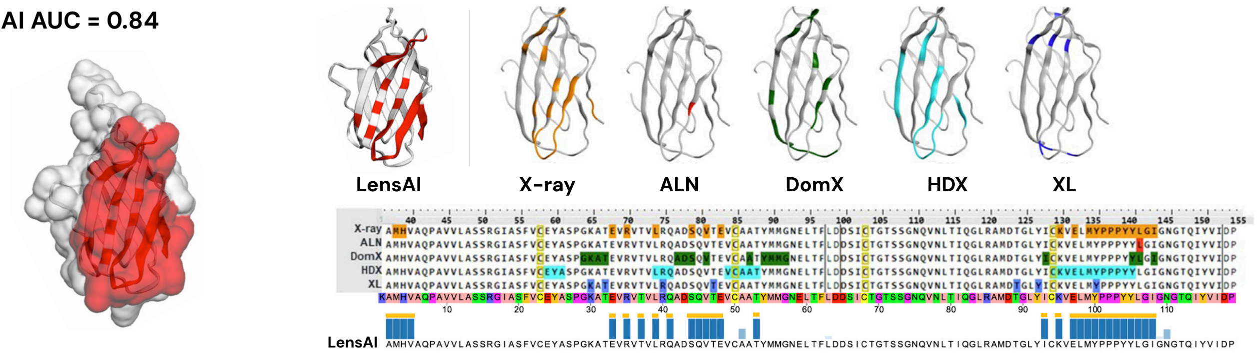
CTLA4 + Tremelimumab

LensAI AUC = 0.83



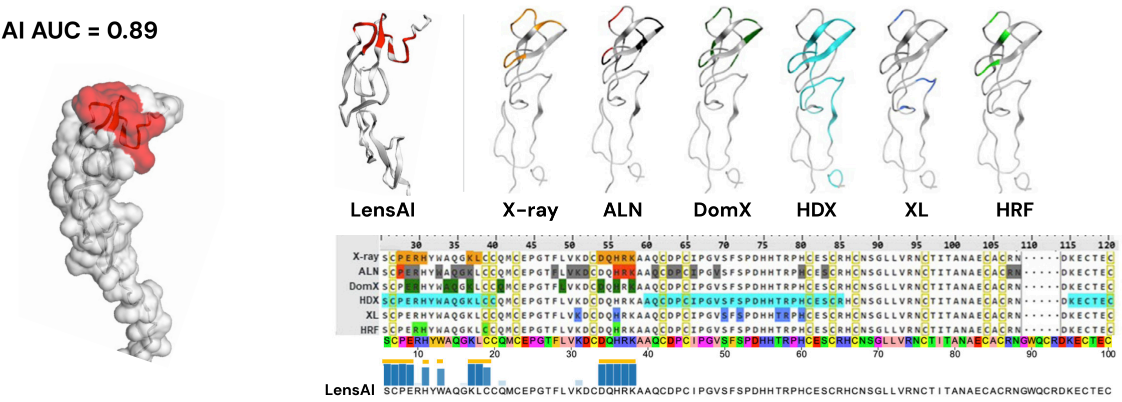
CTLA4 + Ipilimumab

LensAI AUC = 0.84



CD27 + MK-5890

LensAI AUC = 0.89



Method:

The epitope identified by x-ray crystallography is set as ground truth. The following standard metrics are used to quantify epitope prediction accuracy of the different methods:

The True Positive Rate:*

$$TPR = \frac{TP}{(TP+FN)}$$

measures the proportion of residues being part of the true epitope that are correctly identified

The False Positive Rate:

$$FPR = \frac{FP}{(FP+TN)}$$

is the proportion of residues not being part of the true epitope that are incorrectly predicted as part of it

Precision:

$$TP = \frac{TP}{(TP+FP)}$$

measures how many of the residues predicted to be part of the epitope are correctly predicted

*(Recall or Sensitivity)

AUC (Area Under the Curve) plots the True Positive Rate against the False Positive Rate. Measures the ability to distinguish epitopes from non-epitopes. **AUC = 1 means perfect prediction. AUC = 0.5 means no better than random guessing.**

Implementation:

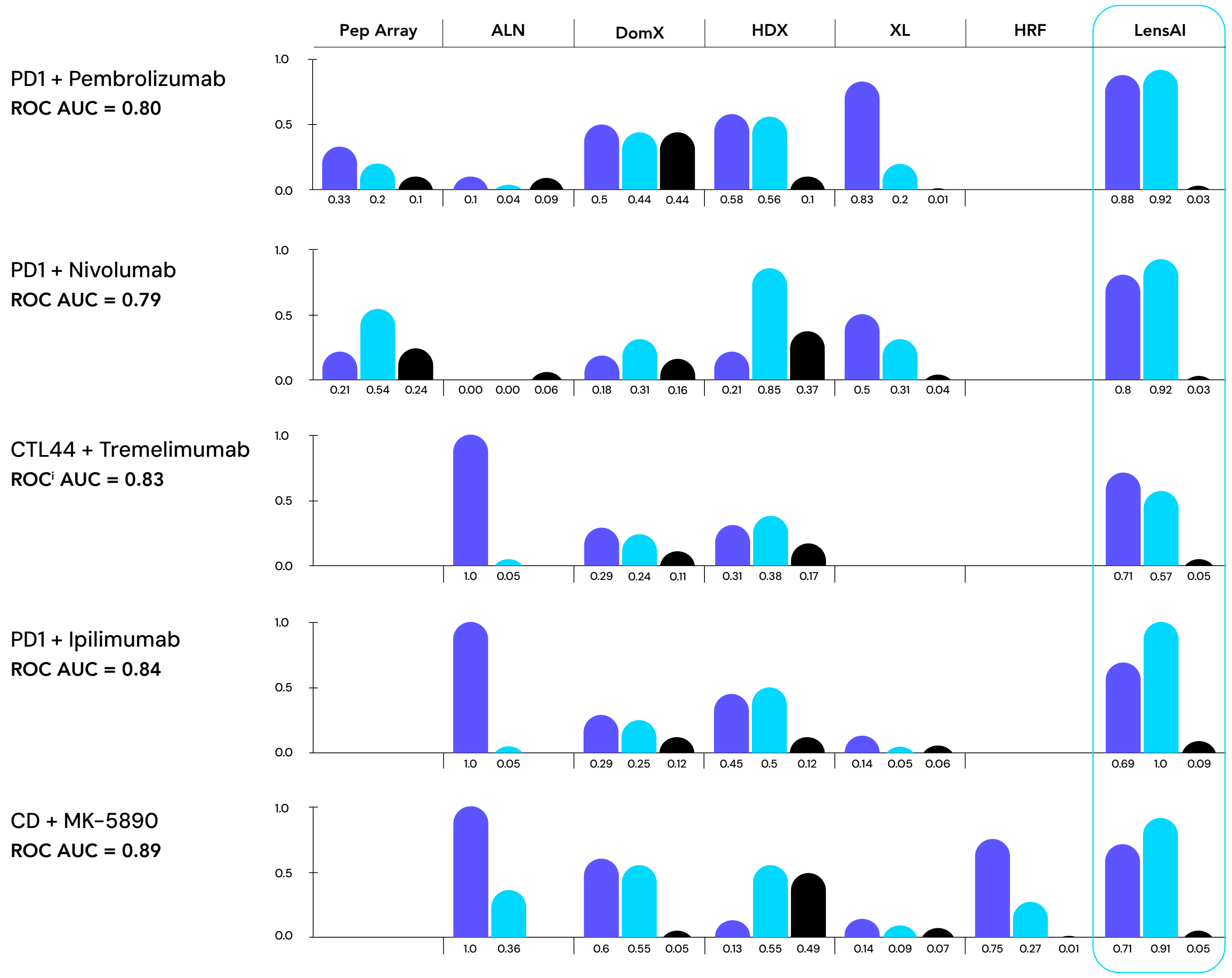
LensAI Epitope Mapping overcomes the limitations of traditional methods—delivering early-stage, high-throughput predictions from sequence alone, with greater speed, scalability, and flexibility across target types.

	LensAI	X-ray Crystallography	Pep Array	ALN	DomX	HDX	XL	HRF
Input	✓ Sequences	Physical Material	Physical Material	Physical Material	Physical Material	Physical Material	Physical Material	Physical Material
Workflow Stage	✓ Flexible – all stages	Late	Early-Mid	Late	Mid	Late	Late	Late
Timeline	✓ Hours/Days	Months	Weeks	Weeks	Weeks	Weeks	Weeks	Weeks
High-Throughput	✓ Yes	No	Yes	No	Partial	Partial	Partial	No
Target Types	✓ Most	Limited	Limited	Limited	Limited	Limited	Limited	Limited

Comparison:

LensAI clearly outperforms all wet-lab based methods and shows a near x-ray crystallography performance. LensAI can accurately identify true epitope residues (high recall or sensitivity) while minimizing wrong predictions (high precision and low FPR).

A well-performing classifier for epitope mapping should accurately identify true epitope residues (**high** recall) while minimizing wrong predictions (**high** precision and **low** FPR).



Conclusion:

LensAI™ Epitope Mapping was evaluated against various wet-lab methods, using epitopes determined by x-ray crystallography as the ground truth. LensAI demonstrated superior performance, achieving near x-ray accuracy. It consistently identified true epitope residues with high sensitivity while maintaining high precision and minimizing false positive rates. Unlike traditional methods, LensAI requires only the target and antibody sequences as input and delivers results within hours, enabling high-throughput application. By providing accurate epitope mapping early in the discovery and development workflow, LensAI accelerates decision-making and reduces risk.

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For more information, contact:
info@mindwalkAI.com

mindwalkAI.com