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Distribution of neoantigen-specific T cells across tumour, lymph node, blood and pleural effusion

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Background

- Neoantigens are tumour-specific antigens generated by somatic mutations and represent attractive targets for personalised cancer immunotherapy¹.
- Neoantigen-specific T cells have been identified in tumours and peripheral blood, but their distribution across distinct immune compartments remains poorly understood².
- Tumour-draining lymph nodes (TDLNs) are important sites of antigen presentation and T-cell priming, while pleural effusion (PE) provides a clinically accessible window into the local immune microenvironment in advanced lung cancer^{3,4}.
- Understanding where neoantigen-specific T cells reside and how they are shared between compartments may inform the development and monitoring of neoantigen-based therapies.

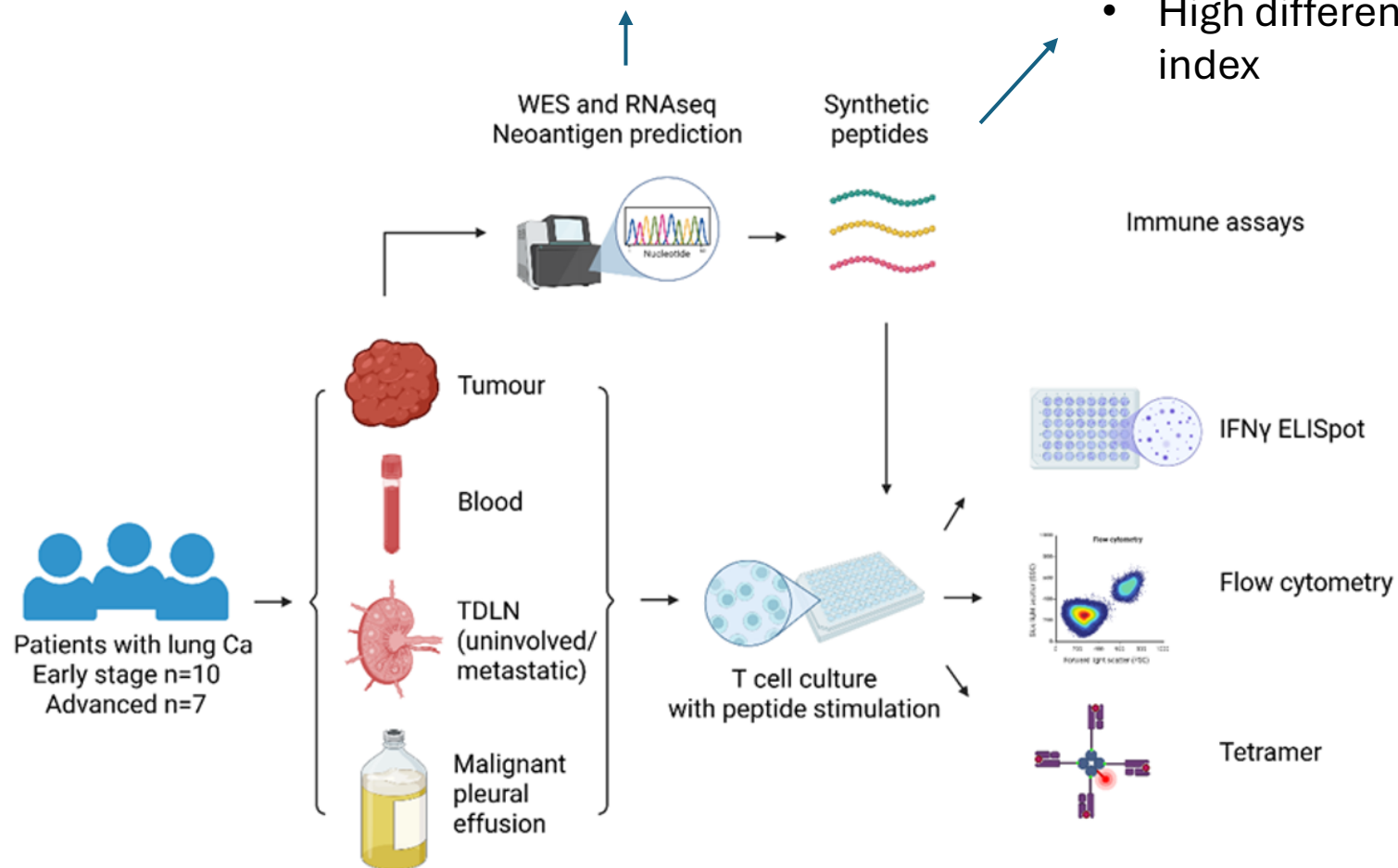
Method

MHC-I neoantigens predicted
using netMHCpan 4.1

- predicted binding affinity $IC_{50} \leq 500$ nM and/or EL rank ≤ 2

Prioritised neoantigens

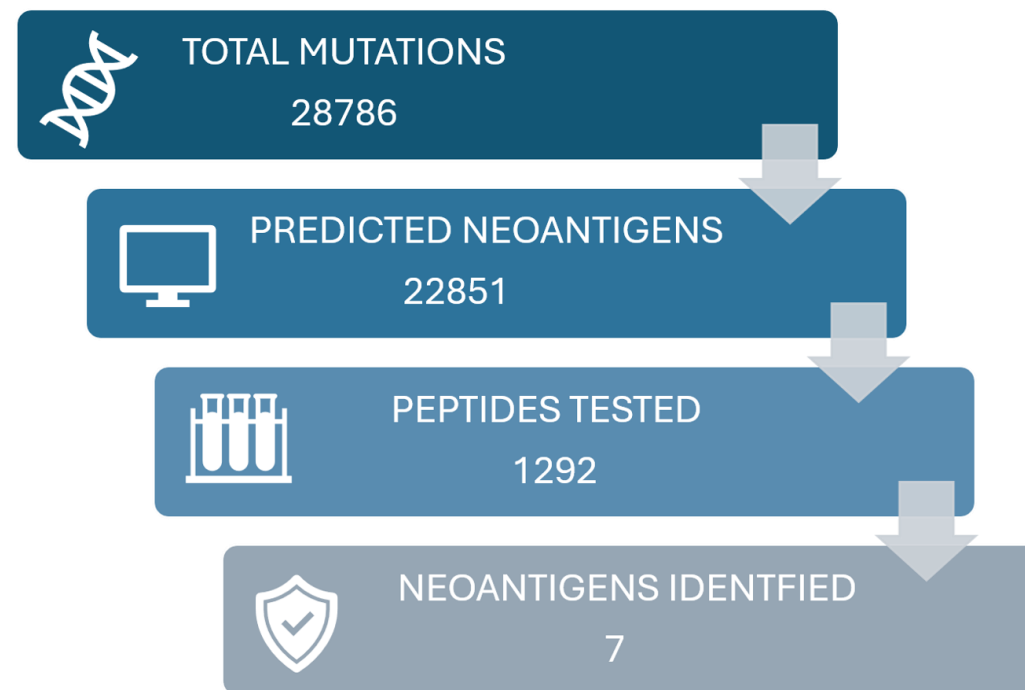
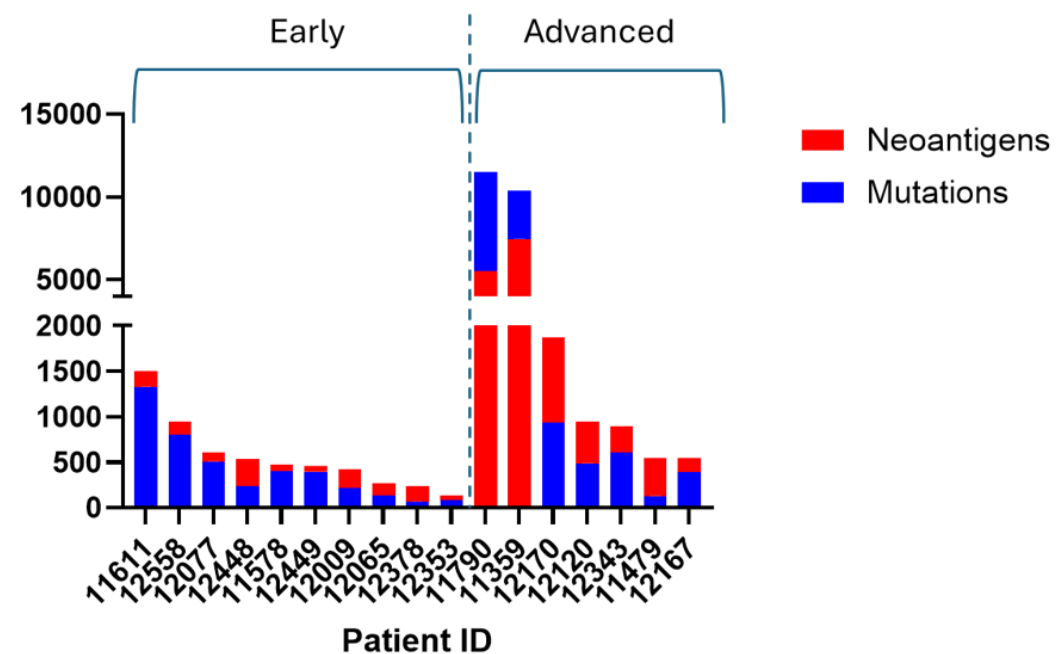
- predicted binding affinity $IC_{50} < 50$ nM and/or EL rank < 0.5
- Gene expression FPKM > 0
- High differential aggretopicity index



Neoantigen prediction and validation

10 early stage and 7 advanced lung cancer patients

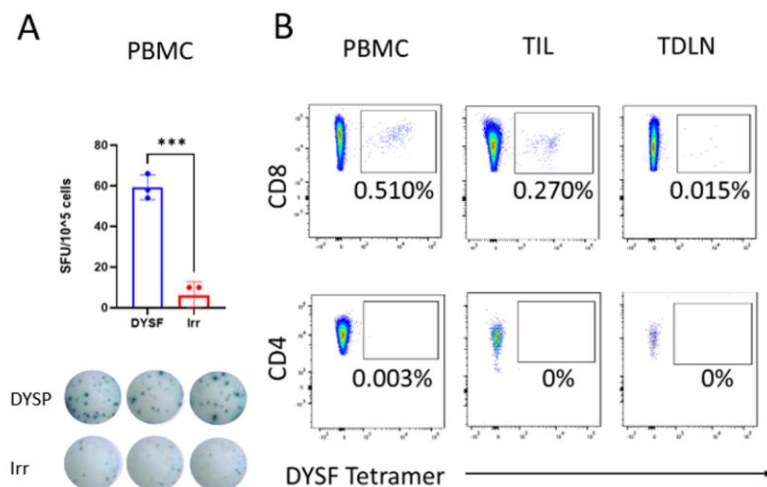
0.77% of predicted neoantigens elicited detectable naturally occurring T-cell responses.



Neoantigen immune response is multicompartmental

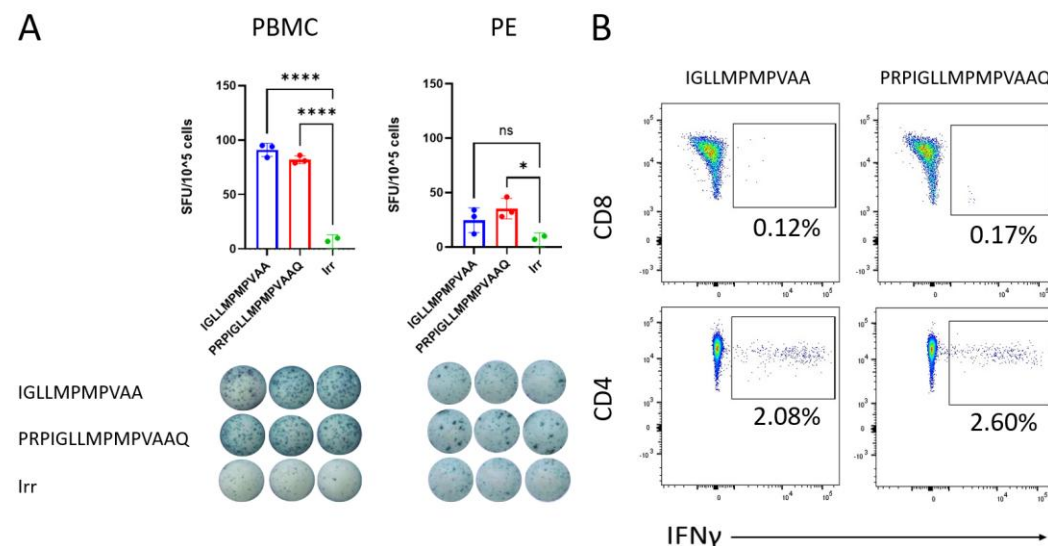
Early stage NSCLC

- A) Detection of IFN γ -secreting T cells against DYSPT^{498C} neoantigen by ELISpot in PBMC.
- B) DYSPT^{498C} tetramer positivity in PBMC, TIL and TDLN CD8⁺ T cells.



Advanced stage SCLC

- A) Detection of IFN γ -secreting T cells against TTBK1^{R1163L} neoantigen by ELISpot in PBMC and PE
- B) Intracellular IFN γ staining confirms a CD4⁺ T-cell response.



Easy to access compartments such as blood and PE can be used to detect and monitor neoantigen-specific T cells.
A single immune assay may underestimate the breadth of endogenous neoantigen-specific T-cell immunity.

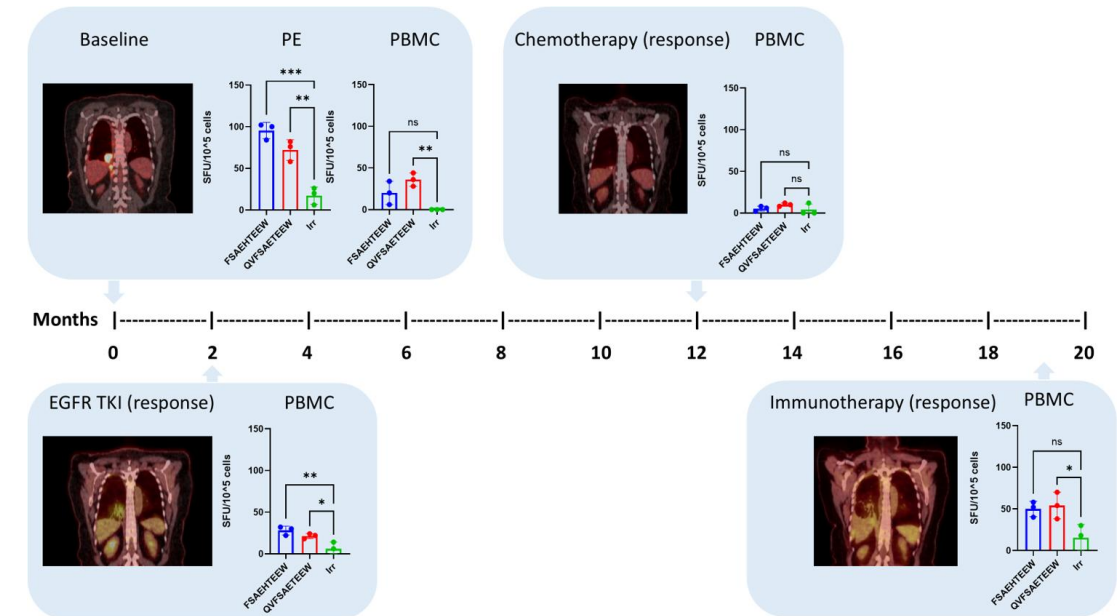
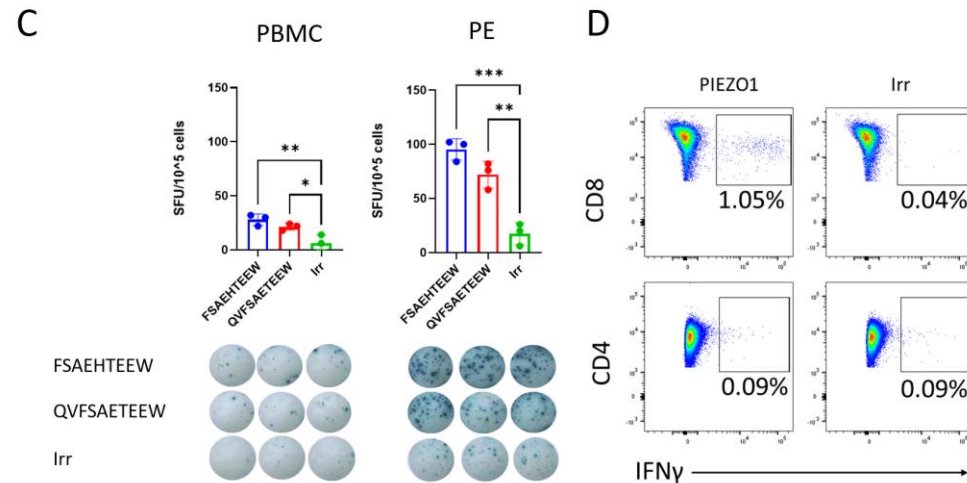
Temporal and spatial detection of neoantigen specific T cells

Advanced stage NSCLC

C) Detection of IFN γ -secreting T cells against PIEZO1^{R1120H} neoantigens by ELISpot in PBMC and PE.

D) Intracellular IFN γ staining and flow cytometry, confirming a CD8⁺ T-cell response.

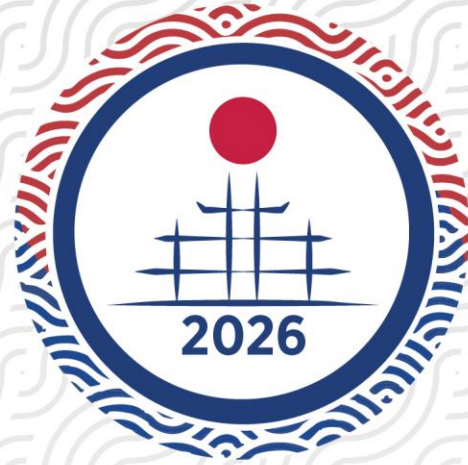
Longitudinal IFN γ T-cell response using ELISpot against PIEZO1^{R1120H} neoantigen during treatment with EGFR TKI, chemotherapy and ICPB over 19 months.



Different immune compartments and longitudinal tracking can improve our understanding of the **spatial and temporal dynamics** of anti-tumour immunity.

References

1. Gupta, R. G., et al. (2021). "Exploiting Tumor Neoantigens to Target Cancer Evolution: Current Challenges and Promising Therapeutic Approaches." Cancer Discov **11(5): 1024–1039.**
2. Duhen, T., et al. (2018). "Co-expression of CD39 and CD103 identifies tumor-reactive CD8 T cells in human solid tumors." Nature communications **9(1): 2724–2713.**
3. Chen, D. S. and I. Mellman (2013). "Oncology meets immunology: the cancer-immunity cycle." Immunity **39(1): 1–10.**
4. Morgensztern, D., et al. (2012). "Prognostic Impact of Malignant Pleural Effusion at Presentation in Patients with Metastatic Non–Small-Cell Lung Cancer." Journal of Thoracic Oncology **7(10): 1485–1489.**



Thank You.