Deciphering Clonal Expansion and the Immune Landscape in Non-Small-Cell Lung Cancer Patients to Identify Biomarkers for Immune Checkpoint Blockade Response

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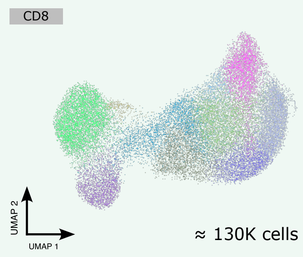
Introduction

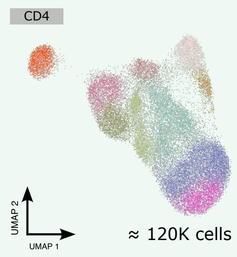
The landscape of Non-Small Cell Lung Cancer (NSCLC) treatment has been transformed by the advent of **immunotherapies** targeting the programmed death 1 (PD-1) and programmed death-ligand 1 (PD-L1) (M. Reck, 2022). The effectiveness of such treatments is limited by the lack of reliable **biomarkers** for predicting therapeutic responses (G. Morad, 2021).
Here, we combined **scRNA** and **deep TCR repertoire sequencing** to explore clonal expansion in 15 NSCLC patients undergoing Immune Checkpoint Blockade (ICB) therapy (anti-PDL1).

1. Differences in T cell composition and activity across responses

Responder (R) = 6; Non-Responder (NR) = 9

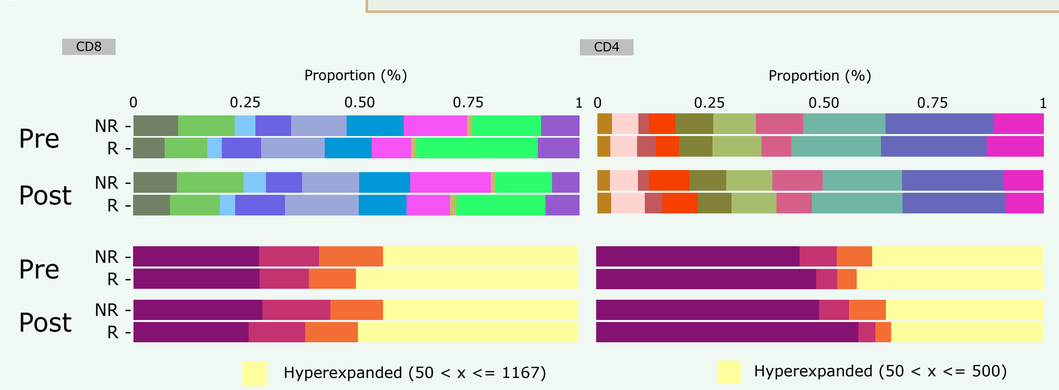
A





- Naive CD8
- Central Memory CD8
- Central Memory Intermediate CD8
- Effector Memory CD8
- TCF7+ Activated Effector Memory CD8
- CX3CR1+ Hobit+ Effector Memory CD8
- Cytotoxic Effector CD8
- TOX+ Effector CD8
- FOS+ Effector CD8
- TCF7+ GZMK+ Effector CD8
- Naive CD8
- Central Memory CD4
- Effector Memory CD4
- Th1
- Th2
- Th17
- Cytotoxic CD4
- HLA+ CD4
- Tregs
- iTREGs

B

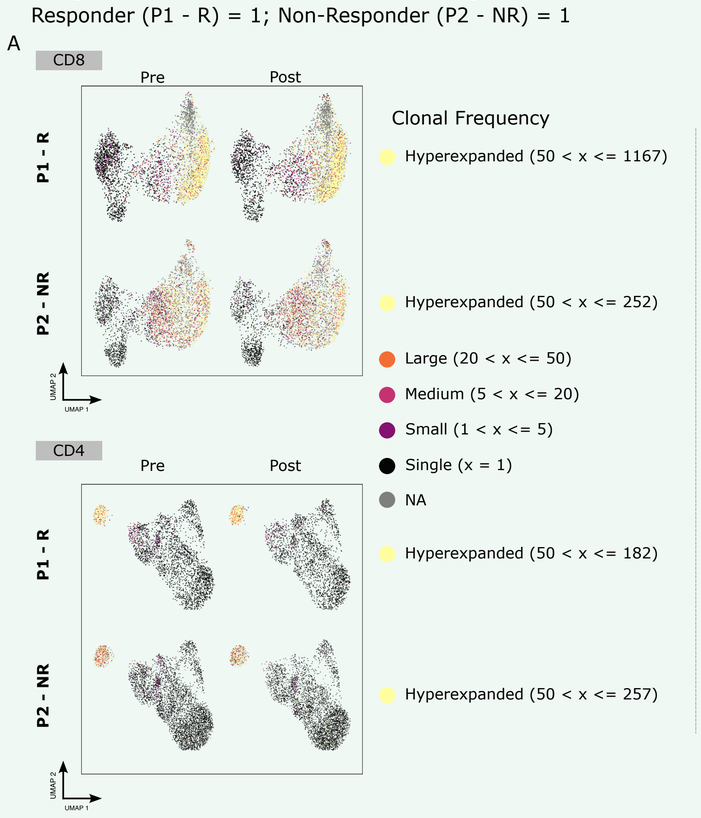


- ↑ Central Memory CD8 in Responders
- ↑ CD8 expansion in Responders Pre-ICB

2. Repertoire features in Responders and Non Responders

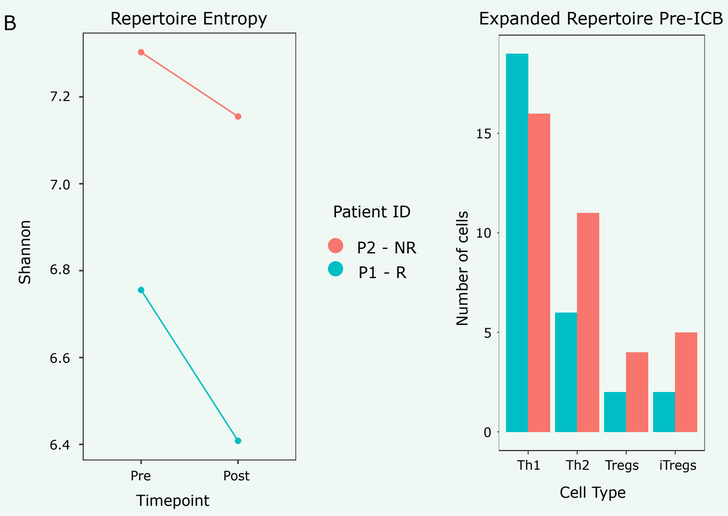
Responder (P1 - R) = 1; Non-Responder (P2 - NR) = 1

A



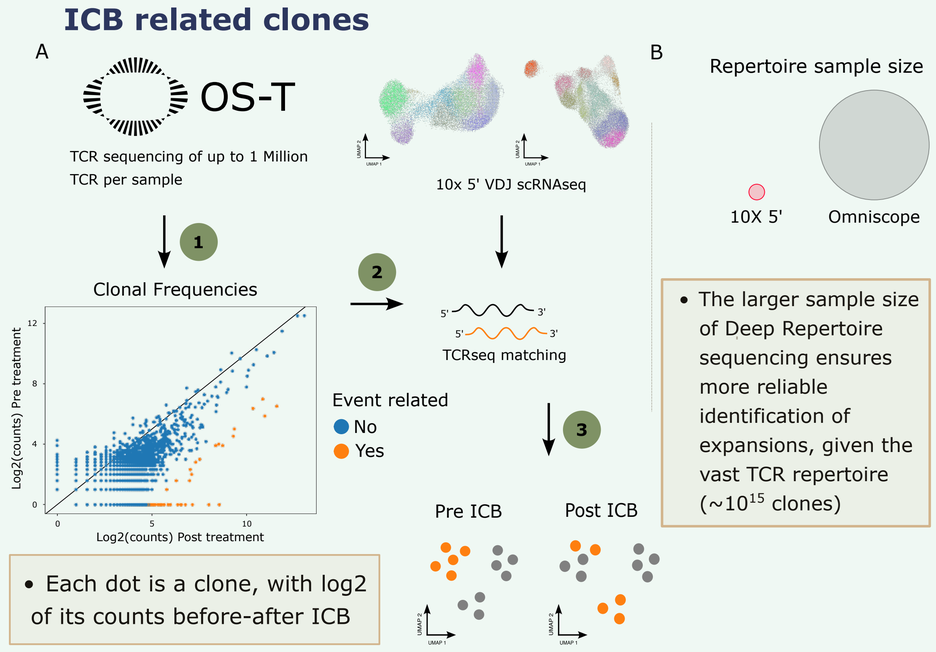
- Localized clonal expansion in Responders

B



- ↑ Repertoire expansion in Responders
- ↑ Type-2 + regulatory activity in Non-Responders

3. Deep Repertoire & phenotypical back-tracing of ICB related clones



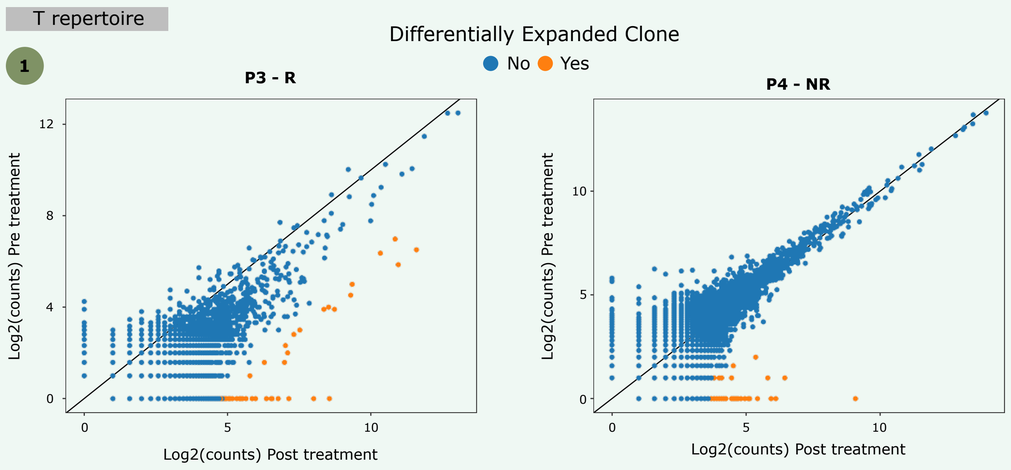
- The larger sample size of Deep Repertoire sequencing ensures more reliable identification of expansions, given the vast TCR repertoire (~10^{15} clones)
- Each dot is a clone, with log2 of its counts before-after ICB

4. Phenotyping of ICB-induced clones

A

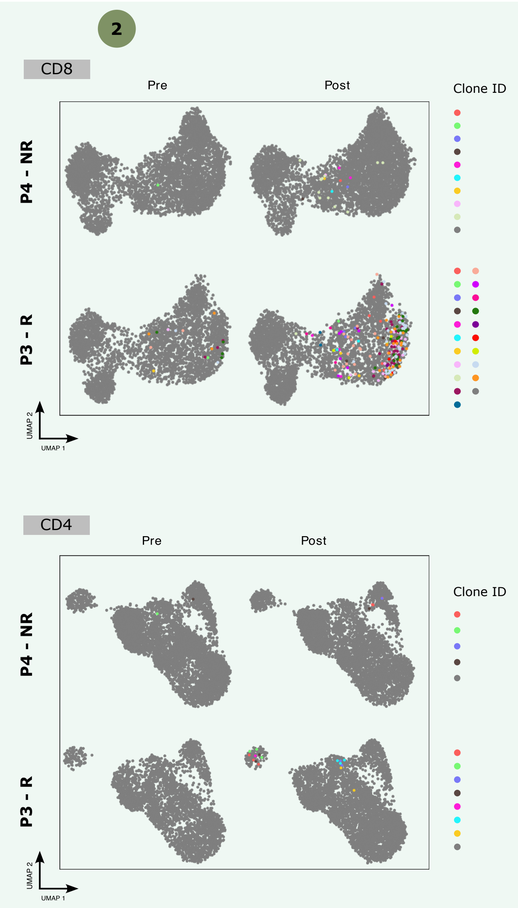
Responder (P3 - R) = 1; Non-Responder (P4 - NR) = 1

T repertoire



- ICB induces differential clonal expansion in Responders

B



C

- High TCF7+ Eff. Mem. CD8 clones
- Switch to CX3CR1+ Hobit+ Eff. Mem. CD8 in Responders
- Tregs expansion in Non Responders
- Cytotoxic CD4 expansion in Responders

Conclusions

- Our data suggest that responders exhibit localized clonal expansion within specific CD4 and CD8 populations, particularly effector memory CD8+ T-cells, compared to non-responders where the clonal expansion exhibit a broader distribution across various populations.
- Some responders show a predominant type 1 immune response, whereas non-responders have an increase in Th2 clones and Tregs expansion.

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