



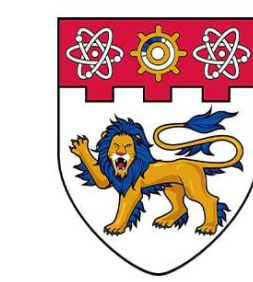
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P3.096: Integrated Spatial and Proteomic Profiling Identifies Metabolic-Immune States Associated with Therapeutic Resistance in NSCLC

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INTRODUCTION

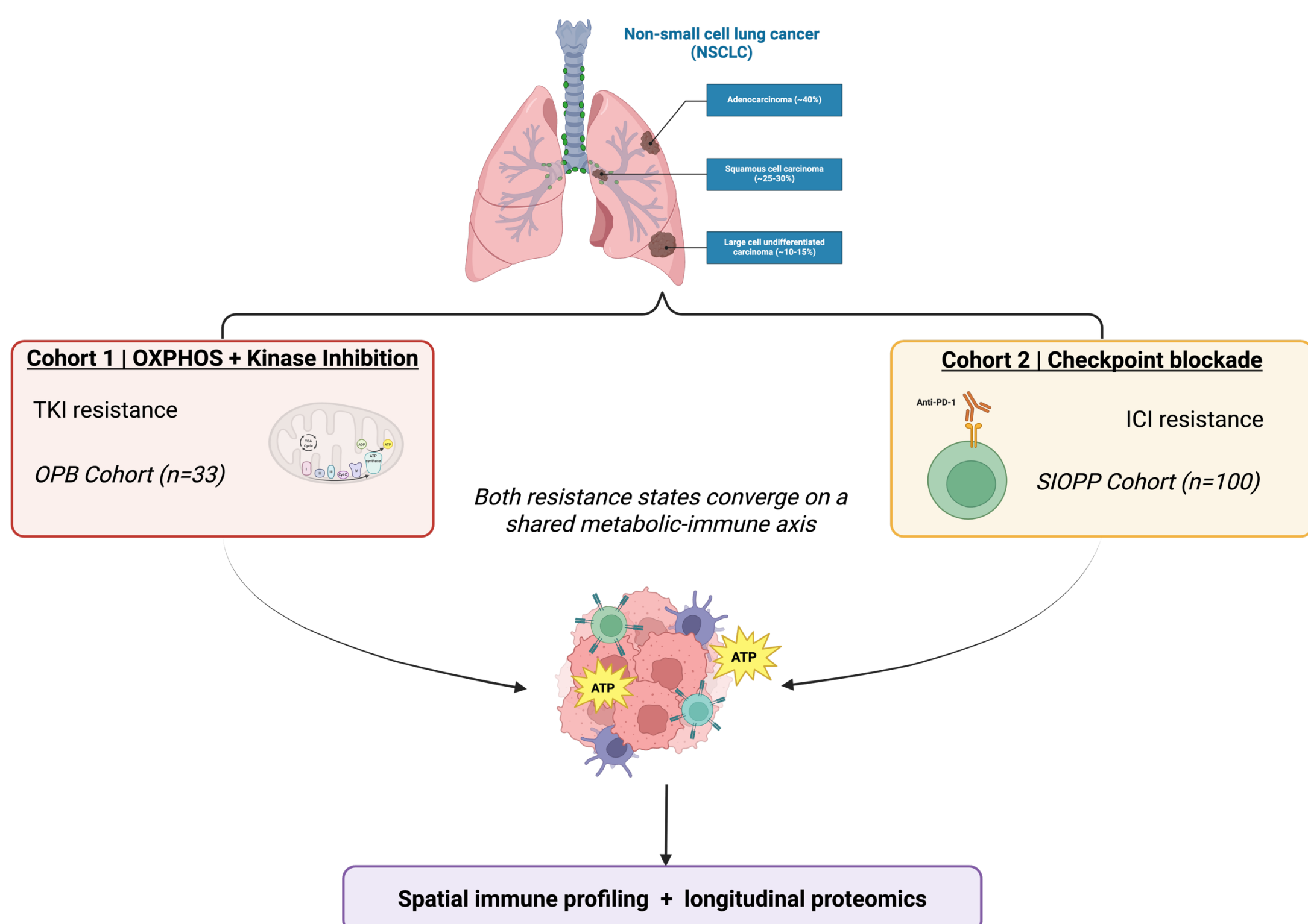
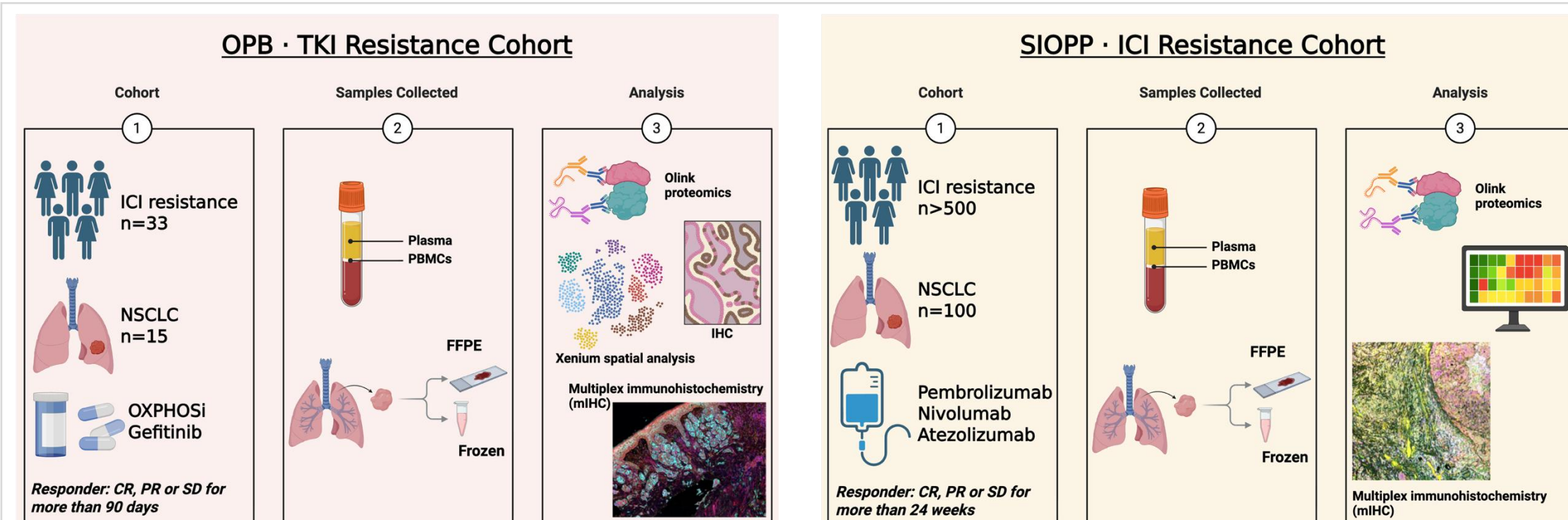


Figure 1. Introduction We hypothesised that resistance states converge on a common metabolic-immune axis. Patients with advanced NSCLC progressing on either OXPHOS-directed kinase inhibition (OPB, n = 33) or checkpoint blockade (SIOPP, n = 100) are compared. Both independent cohorts are analysed in parallel, feeding a single read-out platform combining multiplex immunohistochemistry, serial plasma proteomics and in-situ spatial assays and Xenium transcriptomics in the panel.

METHODOLOGY



Integrated multi-omic analysis of metabolic-immune resistance

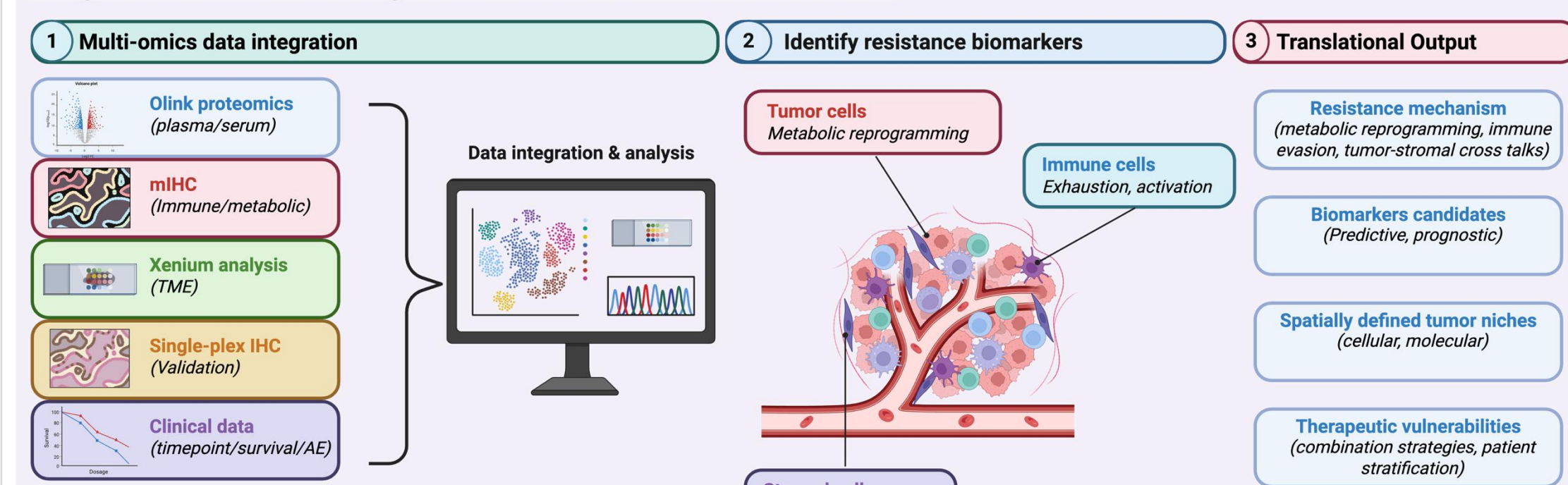


Figure 2. Study design and integrated multi-omic analysis framework. Left: OPB (TKI resistance cohort, n=33 screened, n=15 NSCLC) patients treated with an OXPHOS inhibitor plus gefitinib, stratified by response (CR/PR/SD ≥90 days), with matched sample collection for Olink proteomics, Xenium spatial analysis, and mIHC. Right: SIOPP (ICI resistance cohort, n=500 screened, n=100 NSCLC) patients treated with checkpoint inhibitors (pembrolizumab, nivolumab, atezolizumab), stratified by response (CR/PR/SD ≥24 weeks), with plasma, PBMCs, and FFPE/frozen tumor tissue collected for Olink proteomics and multiplex immunohistochemistry (mIHC). Bottom: Data from both cohorts are integrated for hypotheses testing.

REFERENCES

- Eu JQ, Binte Mohamed Salleh NA, Hirpara J, Chi N, Takaki EO, Tan TZ, Seet JE, Hue SS, Chan SI, Lim DXY, Wang L, Wong RTX, Ali A, Lim YC, Goh BC, Kong LR, Pervaiz S, Wong A. Mitochondrial translocation of MDK2 and TFAM reprograms metabolism in treatment-refractory cancers. *NPI Precis Oncol*. 2026 Apr 16;10(1):270. doi: 10.1038/s41698-025-01257-1. PMID: 41991605; PMCID: PMC13357733.
- Hirpara J, Eu JQ, Tan JKM, Wong AL, Clement MW, Kong LR, Chi N, Tsunoda T, Qu J, Goh BC, Pervaiz S. Metabolic reprogramming of oncogene-addicted cancer cells to OXPHOS as a mechanism of drug resistance. *Redox Biol*. 2019 Jul;25:101076. doi: 10.1016/j.redox.2018.101076. Epub 2018 Dec 17. PMID: 30642723; PMCID: PMC6859574.
- Huang Y, Zhao JJ, Soon YY, Kee A, Tay SH, Aminkeng F, Ang Y, Wong ASC, Bhargava LD, Goh BC, Soo RA. Factors Predictive of Primary Resistance to Immune Checkpoint Inhibitors in Patients with Advanced Non-Small Cell Lung Cancer. *Cancers (Basel)*. 2023 May 12;15(10):2733. doi: 10.3390/cancers15102733. PMID: 37345072; PMCID: PMC10216169.

RESULTS

Figure 3. Responders have elevated expression of ETC Complex I subunit and IFN γ in baseline blood plasma

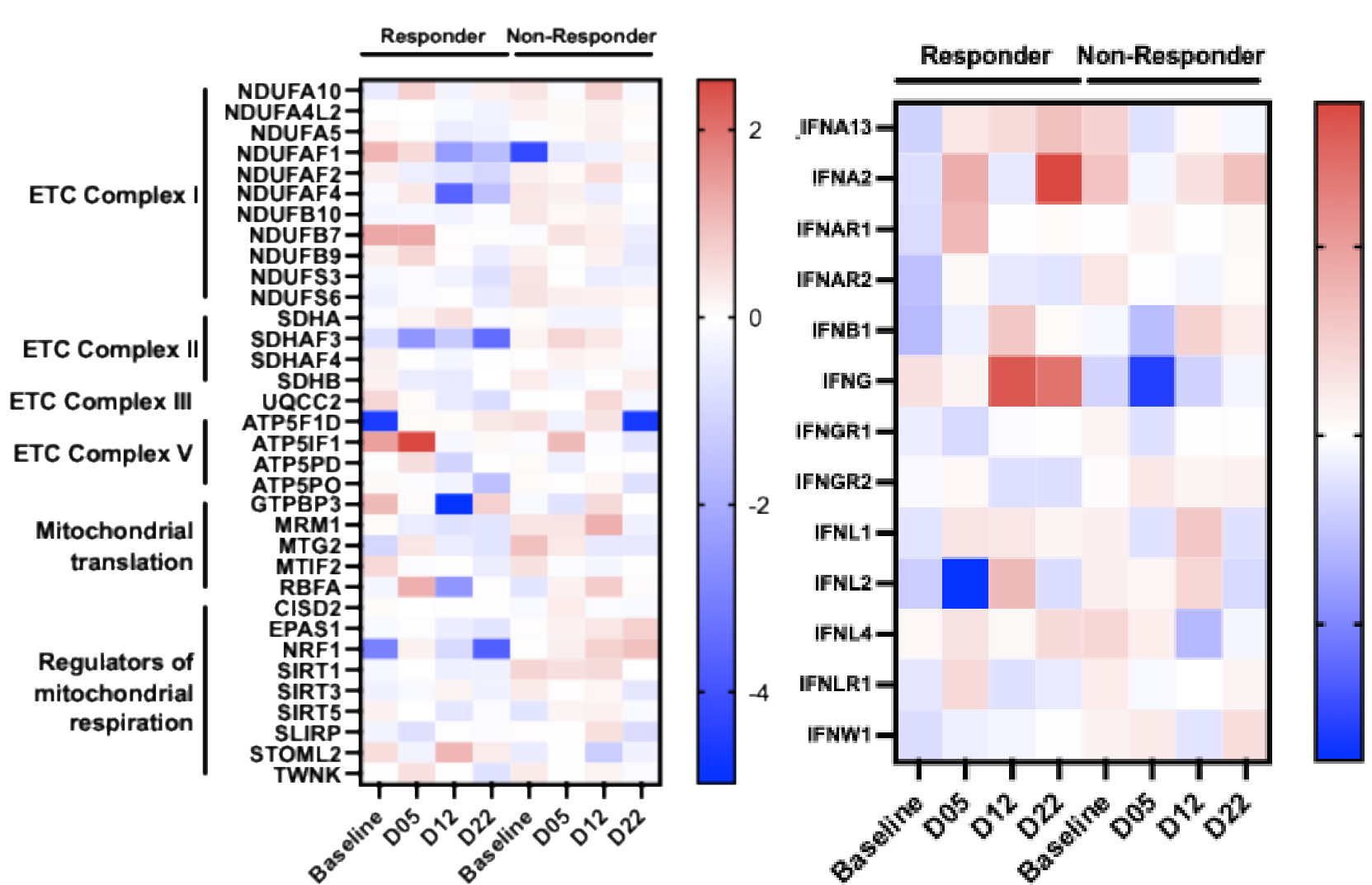


Figure 3. Responders have elevated expression of ETC Complex I subunit and IFN γ in baseline blood plasma. Serial plasma Olink proteomics, OPB cohort (baseline, D05, D12, D22). Left/middle: heatmaps of ETC Complex I-V/mitochondrial-regulator and interferon-pathway protein levels, responders vs non-responders. Right: volcano plot of non-responder vs responder (NR-R) fold-change at baseline; NDUFA1 and PSMA2 were reduced, and CTSL3P, MAGEC2, RIPK4, RN4F4 elevated, in non-responders (p<0.05). Elevated IFN γ reflects a more inflamed baseline immune state, consistent with the metabolic-immune axis hypothesis.

Figure 4. Single-plex IHC did not observe significant changes in protein expression post-treatment

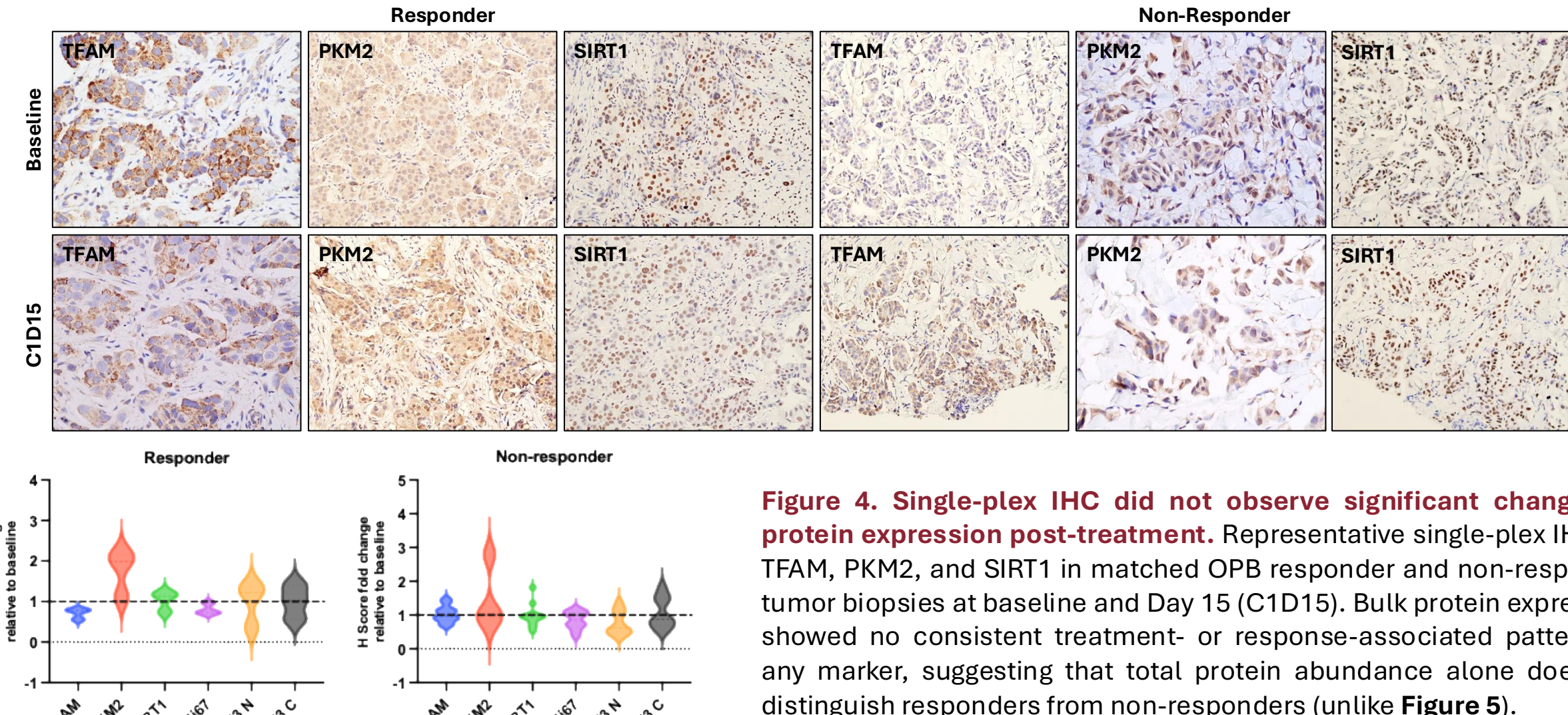


Figure 4. Single-plex IHC did not observe significant changes in protein expression post-treatment. Representative single-plex IHC for TFAM, PKM2, and SIRT1 in matched OPB responder and non-responder tumor biopsies at baseline and Day 15 (D15). Bulk protein expression showed no consistent treatment- or response-associated pattern for any marker, suggesting that total protein abundance alone does not distinguish responders from non-responders (unlike Figure 5).

Figure 5. TFAM mitochondrial localization positively correlates to OXPHOSi + TKI response

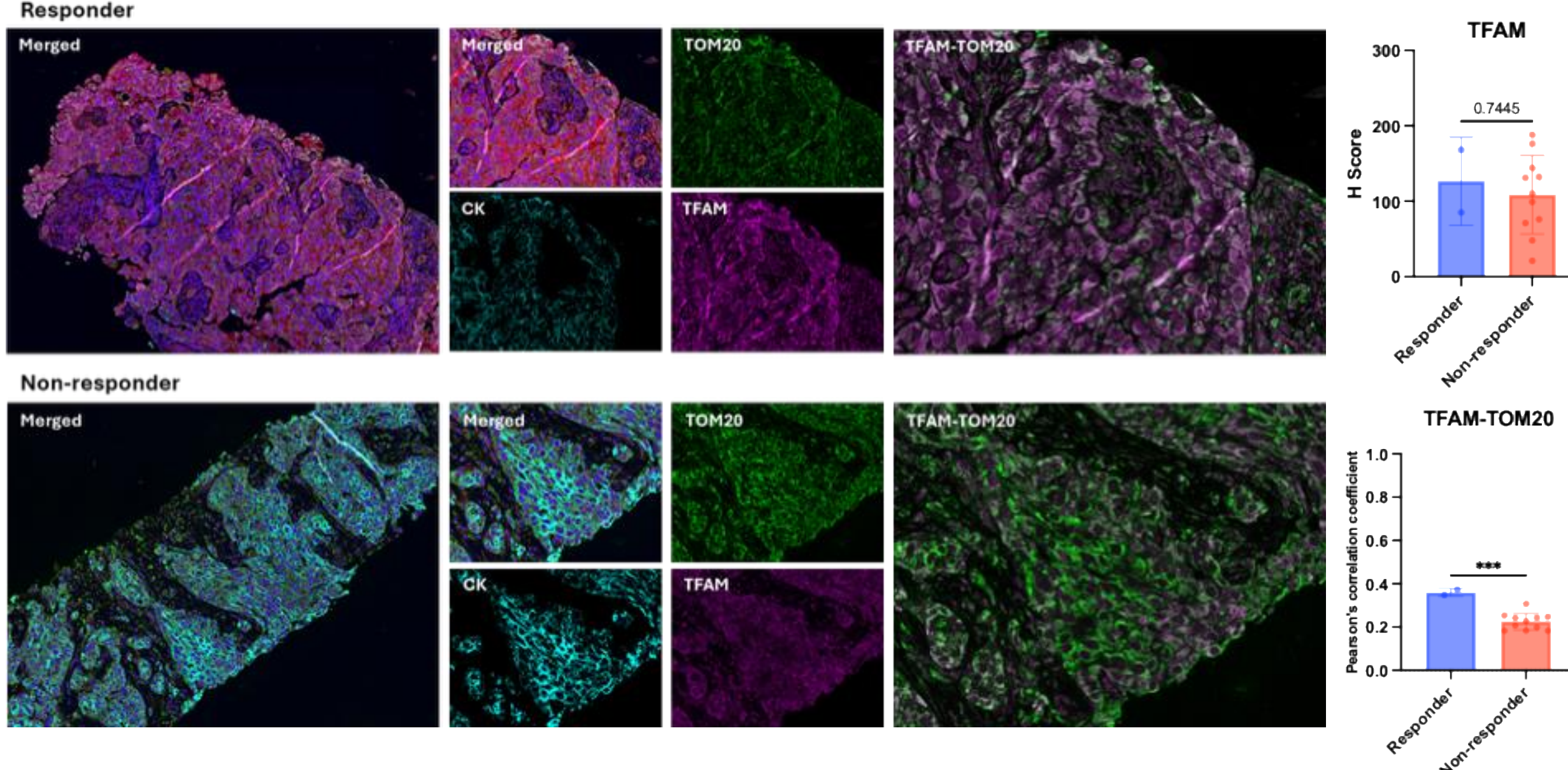


Figure 5. TFAM mitochondrial localization positively correlates to OXPHOSi + TKI response. Multiplex immunohistochemistry (mIHC) for pan-cytokeratin (CK), TOM20, and TFAM in matched tumor biopsies from a responder and non-responder from OPB cohort. Representative whole-slide merge, zoomed single-channel images, and a TFAM-TOM20 co-localization overlay (white/grey = overlap) are shown. Spatial co-localization of TFAM with TOM20 was visibly greater in the responder, quantified by Pearson's correlation coefficient, suggesting that TFAM mitochondrial localization and not the protein abundance is significantly higher in responders (***p<0.001).

Figure 6. Spatial transcriptomics reveals diverse immune composition that correlates with response to therapy

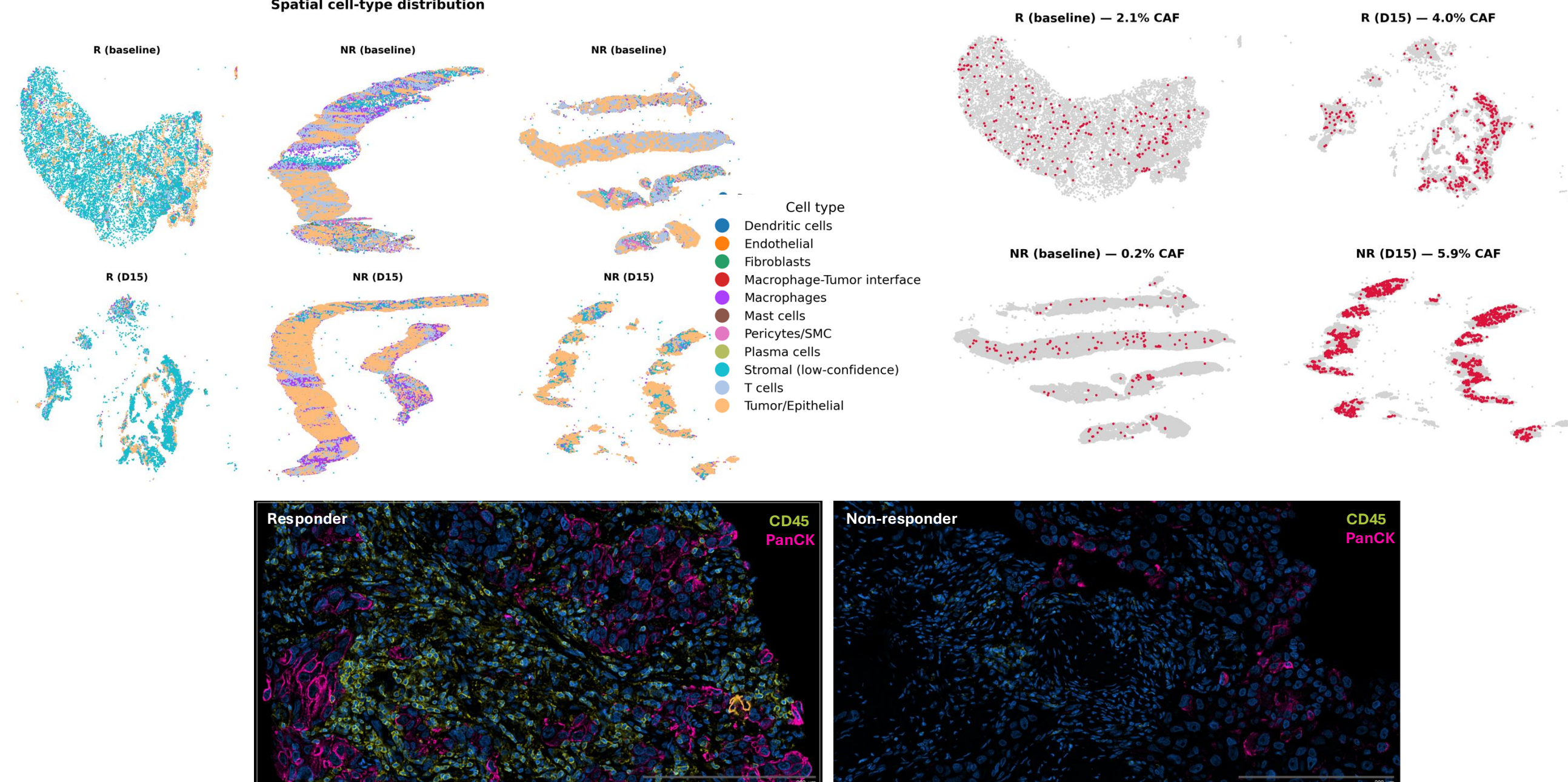


Figure 6. Spatial transcriptomics reveals diverse immune composition that correlates with response to therapy. OPB cohort (responder R; non-responders NR; baseline and D15). Left, top: spatial cell-type distribution (totalVI joint RNA+protein clustering); the "Stromal (low-confidence)" population in R reflects a technical QC artifact (lower tissue transcript density). Right, top: core fibroblast/CAF subcluster as % of cells, baseline vs D15, with representative spatial maps (red). Center, bottom: representative CD45/pan-cytokeratin (PanCK) images showing denser immune infiltrate in the responder vs non-responder. n=1 responder, n=2 non-responders;

Figure 7. Immune infiltration alone does not stratify ICI response

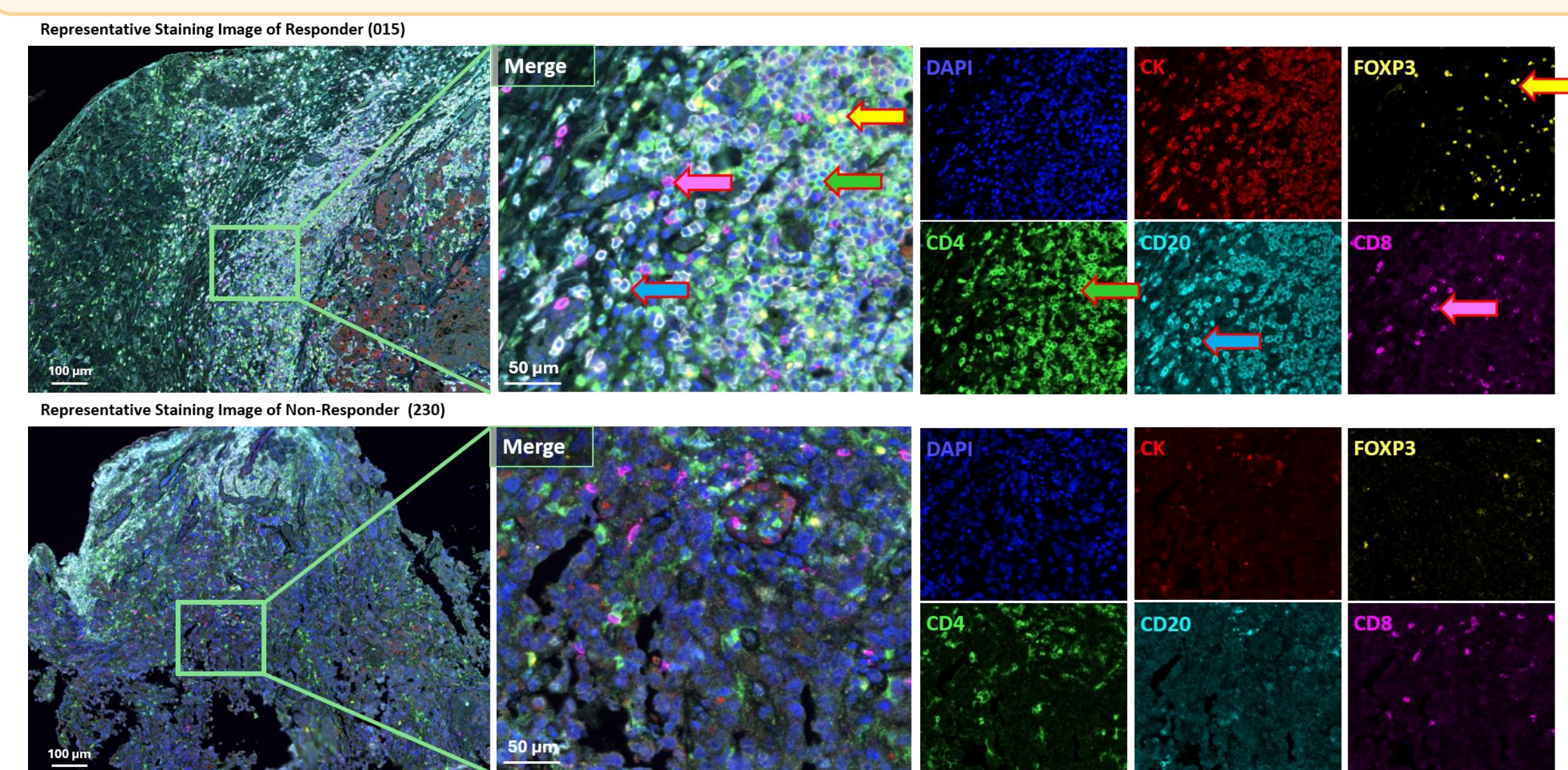


Figure 7. Immune infiltration alone does not stratify ICI response as a "immune-hot" phenotype is confined to a subset of responders. mIHC (DAPI, CK, FOXP3, CD4, CD20, CD8) of SIOPP tumour biopsies from responders (n=19) and non-responders (n=4). Representative staining from a responder (patient 015) and a non-responder (patient 230); whole-slide, zoomed-merge, and single-channel images shown, with arrows marking FOXP3+, CD4+, CD8+, and CD20+ cells. High total infiltration was restricted to a subset of responders, with the majority of responders showing counts comparable to non-responders, suggesting other underlying mechanisms shaping response towards ICI treatment.

Figure 8. Increase in immune checkpoint, ECM and metabolic markers in responders after treatment

