

expression was highly variable and distinct PD-L1 driven immune phenotypes were identified based on the PD-L1 intensity on both tumor and immune cells, the distance between non-exhausted T-cell subsets (i.e. PD-1 and CTLA-4 expression on CD3⁺CD8⁺ cytotoxic T-cells, CD3⁺CD4⁺ T-helper cells, CD3⁺CD4⁺FOXP3⁺ regulatory T-cells) and tumor cells as well as macrophage/(DC) subtypes. In breast cancer, the PD-L1 fluorescence intensity on tumor cells showed a significantly higher predictive performance for overall survival with an area under receiver operating curves (AUC) of 0.72 ($p < 0.0001$) than the percentage of PD-L1⁺ tumor cells (AUC: 0.54). In PD-L1 positive as well as negative breast cancers a close spatial relationship between T-cell subsets (CD3⁺CD4⁺CD8⁺FOXP3⁺PD-1⁺CTLA-4⁺) and Macrophage/DC subsets (CD68⁺CD163⁺CD11c⁺iNOS) was found prognostic relevant ($p < 0.0001$).

Conclusions In conclusion, multiplex immunofluorescence PD-L1 assessment provides cutoff-free/continuous PD-L1 data which are superior to the conventional percentage of PD-L1⁺ tumor cells and of high prognostic relevance. The combined analysis of spatial PD-L1/PD-1 data and more than 20 different immune cell subtypes of the immune tumor microenvironment revealed distinct PD-L1 immune phenotypes.

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P02.04 TISSUE-INFILTRATING TH9 CELLS IN HUMAN ENDOMETRIAL CANCER

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Background Endometrial cancer (EC) is a hormone-related carcinoma with increased morbidity among female patients of all backgrounds. The immune microenvironment of EC is uncertain.

Materials and Methods 102 patients were recruited in the present study. 90 postoperative specimens from the patients were analyzed by immunohistochemistry. The leukocyte landscape of endometrial cancer was mapped using high-dimensional single-cell profiling (CyTOF) for 12 patients.



Abstract P02.04 Figure 1

Results NK cells, MDMs, and neutrophils were enriched in adjacent normal tissue. CCR5+CD38+ PD1+Th9 cells were enriched in the invasive margin. Additionally, PD1+ESR_{neg} T cells and Siglec1+CCR5+CD40+ESR^{hi} macrophage were infiltrated in the tumors.

Conclusions Immunological landscape of EC might shed light on new immunotherapeutic approach.

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P02.05 COMPREHENSIVE PROFILING OF TUMOR HETEROGENEITY AND ITS MICROENVIRONMENT IN ADVANCED NON-SMALL CELL LUNG CANCER AT SINGLE CELL RESOLUTION

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Background Lung cancer is a highly heterogeneous disease. Cancer cells and cells within the tumor microenvironment together determine disease progression, as well as response to or escape from treatment.

Materials and Methods To map the cell type-specific transcriptome landscape of cancer cells and their tumor microenvironment in advanced non-small cell lung cancer (NSCLC), we analyzed 42 tissue biopsy samples from stage III/IV NSCLC patients by single cell RNA sequencing and presented the large scale, single cell resolution profiles of advanced NSCLCs.

Results In addition to cell types described in previous single cell studies of early stage lung cancer, we were able to identify new cell types such as follicular dendritic cells and T helper 17 cells. Tumors from different patients display large heterogeneity in cellular composition, chromosomal structure, developmental trajectory, intercellular signaling network and phenotype dominance. Our study also revealed a correlation of tumor heterogeneity with tumor associated neutrophils, which might help to shed light on their function in NSCLC.

Conclusions This study presented first-time the tumor heterogeneity and tumor microenvironment profile from late-stage, largely untreated NSCLC patients, and shed light on possible treatment regimes.

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P02.06 SEMI-AUTOMATED VALIDATION AND QUANTIFICATION OF CTLA-4 IN 90 DIFFERENT TUMOR ENTITIES USING MULTIPLE ANTIBODIES AND ARTIFICIAL INTELLIGENCE

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Background CTLA-4 is an inhibitory immune checkpoint receptor and a negative regulator of anti-tumor T-cell