**PRECISE SPATIAL MULTIPLEXING FOR IMMUNE
PROFILING IN NON-SMALL CELL LUNG CANCER FFPE
SAMPLES WITH CHIPCYTOMETRY**

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Background Emergent data indicate that highly multiplexed spatial biomarker analysis has the potential to advance precision medicine in immuno-oncology and inform the discovery of novel biomarkers.

Methods Here we present the analysis of clinical FFPE samples from non-small cell lung cancer patients using ChipCytometry, a novel precise spatial multiplexing technology which combines iterative immuno-fluorescent staining with high-dynamic range imaging to facilitate quantitative phenotyping with single-cell resolution. Standard FCS files are generated from multichannel OME-TIFF images, enabling identification of cellular phenotypes via flow cytometry-like hierarchical gating. In this study, a 27-plex assay was used to identify and quantify more than 30 cellular phenotypes and subtypes in FFPE samples.

Results The results show precise expression levels for each marker in the assay in each individual cell in the sample, maintaining spatial information about each cell. Spatial analysis of the samples reveals quantifiable heterogeneity of immune cell infiltration within the tumor samples, demonstrating the utility of the ChipCytometry platform for the in-depth immune profiling in clinical samples.

Conclusions N/A

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