Spatial proteomic and transcriptomic biomarkers of response to immune checkpoint inhibitors in operable lung cancer

Jonathan Stauber, Corinne Ramos*, Adele Ponzoni, Amandine Gerstenberg. Imabiotec, Loos, France

Background Non-small cell lung cancer (NSCLC) accounts for about 85% of all lung cancers. There is a strong rationale for incorporating immunotherapy into the treatment of early-stage NSCLC, given the breakthrough results with PD-1 checkpoint inhibitors in advanced-stage NSCLC. How immunotherapy should be implemented in patients who are operable is still unclear. Most of the efforts so far to identify clinically useful biomarkers do not preserve spatial information and leave us blind to the critical source of information revealed in the cell-to-cell biology of the tumor microenvironment (TME). In order to overcome these limitations, we used spatial biomarkers assays that preserve this critical information about which cells are influencing treatment response.

Methods Frozen sections from retrospectively collected surgically resected NSCLC (adenocarcinoma and squamous cell carcinoma) tumors treated with adjuvant pembrolizumab therapy were used. Patients were classified in two groups according to their Objective Response Rate (ORR): Complete Response (CR) and Progression Disease (PD) for spatial transcriptomic and proteomics assays. The statistical analysis was performed through the GeoMx® DSP analysis suite. Cell deconvolution using the SpatialDecon® algorithm (Nanostring®) was then used to estimate the cell-type abundance in the spatially-resolved region of interest. Results were validated with single cell proteomic spatial analysis using proprietary workflow to identify which cells are influencing the treatment response and how they are spatially distributed relative to each other.

Results A higher expression of the drug targets, PD1 (PDCD1) and PD-L1 (CD274), and genes related to T lymphocytes cytotoxicity (GZMB, CD8a) and activation (CD44, CD27, TNFRS9) were detected in the tumor microenvironment of the responder patient. The analysis of the non-responder patients highlighted the overexpression of inhibitory ligands CD86 and B7H3 (CD276). Interestingly, TIGIT, CTLA4 and TIM-3 were significantly overexpressed on the surface of the CD8a+ T cells. These results were validated by investigating the drug targets and immunosuppressive cells in the tumor microenvironment of patient samples that did not respond to immunotherapy.

Conclusions These findings highlight the relevance of considering a set of spatial biomarkers involved in immune suppression pathways to obtain a comprehensive portrait of the tumor microenvironment for personalized therapy selection. Our results suggest that for patients who did not respond to monotherapy, it would have been preferable to resort to a combined immune checkpoint inhibitors treatment strategy, aimed at the complete inhibition of all the immune-suppressive pathways.