acid and MTOR metabolic pathways as a potential novel therapeutic targets for complementary therapy to restore immune function in melanoma and SCC patients.

Ethics Approval This study was performed under an IRB approved protocol.

P853 SINGLE CELL TRANSCRIPTOME ANALYSIS IDENTIFIES UNIQUE FEATURES IN CIRCULATING CD8+ T CELLS THAT CAN PREDICT IMMUNOTHERAPY RESPONSE IN MELANOMA PATIENTS

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10.1136/LBA2019.7

Background Immune checkpoint blockade (ICB) has greatly advanced the treatment of melanoma. A key component of ICB is the stimulation of CD8+ T cells in the tumor. However, ICB therapy only benefits a subset of patients and a reliable prediction method that does not require invasive biopsies is still a major challenge in the field.

Methods We conducted a set of comprehensive single-cell transcriptomic analyses of CD8+ T cells in the peripheral blood (mPBL) and tumors (mTIL) from 8 patients with metastatic melanoma.

Results Compared to circulating CD8+ T cells from healthy donors (hPBL), mPBLs contained subsets resembling certain features of mTIL. More importantly, three clusters (2, 6 and 15) were represented in both mPBL and mTIL. Cluster 2 was the major subset of the majority of hPBL, which phenocopied hallmark parameters of resting T cells. Cluster 6 and 15 were uniquely presented in melanoma patients. Cluster 15 had the highest PD-1 levels, with elevated markers of both activation and dysfunction/exhaustion; while Cluster 6 was enriched for 'dormant' cells with overall toned-down transcriptional activity except PPAR signaling, a known suppressor for T cell activation. Interestingly, unlike other mTIL clusters that would classically be defined as exhausted, Cluster 15 exhibited the highest metabolic activity (oxidative-phosphorylation and glycolysis). We further analyzed total sc-transcriptomics using cell trajectory algorithms and identified that these three clusters were the most distinct subtypes of CD8 T cells from each other, representing: resting (cluster 2), metabolically active-dysfunctional (cluster 15), and dormant phenotypes (cluster 6). Further, three unique intracellular programs in melanoma drive the transition of resting CD8+ T cells (cluster 2) to both metabolic/dysfunctional (cluster 15) and dormant states (cluster 6) that are unique to tumor bearing conditions. Based on these high-resolution analyses, we developed original algorithms to build a novel ICB response predictive model using immune-blockade co-expression gene patterns. The model was trained and tested using previously published GEO datasets containing CD8 T cells from anti-PD-1 treated patients and presented an AUC of 0.82, with 92% and 89% accuracy of ICB response in the two datasets.

Conclusions We identified and analyzed unique populations of CD8+ T cells in circulation and tumor using high-resolution single-cell transcriptomics to define the landscape of CD8+ T cell states, revealing critical subsets with shared features in PBLs and TILs. Most importantly, we established an innovative model for ICB response prediction by using peripheral blood lymphocytes.

Ethics Approval This study was performed under an IRB approved protocol.

Checkpoint blockade therapy

P854 CONSTRUCTION OF THE IMMUNE LANDSCAPE OF DURABLE RESPONSE TO CHECKPOINT BLOCKADE THERAPY BY INTEGRATING PUBLICLY AVAILABLE DATASETS

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10.1136/LBA2019.8

Background Immune checkpoint blockade (ICB) has revolutionized cancer treatment. However, long-term benefits are only achieved in a small fraction of patients. Understanding the mechanisms underlying ICB activity is key to improving the efficacy of immunotherapy. A major limitation to uncovering these mechanisms is the limited number of responders within each ICB trial. Integrating data from multiple studies of ICB would help overcome this issue and more reliably define the immune landscape of durable responses. Towards this goal, we formed the TimIOs consortium, comprising researchers from the Society for Immunotherapy of Cancer Sparkathon TimIOs Initiative, the Parker Institute of Cancer Immunotherapy, the University of North Carolina-Chapel Hill, and the Institute for Systems Biology. Together, we aim to improve the understanding of the molecular mechanisms associated with defined outcomes to ICB, by building on our joint and multifaceted expertise in the field of immuno-oncology. To determine the feasibility and relevance of our approach, we have assembled a compendium of publicly available gene expression datasets from clinical trials of ICB. We plan to analyze this data using a previously reported pipeline that

successfully determined main cancer immune-subtypes associated with survival across multiple cancer types in TCGA.[1]

Methods RNA sequencing data from 1092 patients were uniformly reprocessed harmonized, and annotated with predefined clinical parameters. We defined a comprehensive set of immunogenomics features, including immune gene expression signatures associated with treatment outcome,[1,2] estimates of immune cell proportions, metabolic profiles, and T and B cell receptor repertoire, and scored all compendium samples for these features. Elastic net regression models with parameter optimization done via Monte Carlo cross-validation and leave-one-out cross-validation were used to analyze the capacity of an integrated immunogenomics model to predict durable clinical benefit following ICB treatment.

Results Our preliminary analyses confirmed an association between the expression of an IFN-gamma signature in tumor (1) and better outcomes of ICB, highlighting the feasibility of our approach.

Conclusions In line with analysis of pan-cancer TCGA datasets using this strategy (1), we expect to identify analogous immune subtypes characterizing baseline tumors from patients responding to ICB. Furthermore, we expect to find that these immune subtypes will have different importance in the model predicting response and survival. Results of this study will be incorporated into the Cancer Research Institute iAtlas Portal, to facilitate interactive exploration and hypothesis testing.

REFERENCES

1. Thorsson V, Gibbs DL, Brown SD, Wolf D, Bortone DS, Yang T-H O, Porta-Pardo E. Gao GF, Plaisier CL, Eddy JA, *et al*. The Immune Landscape of Cancer. *Immunity* 2018; **48**(4): 812–830.e14. https://doi.org/10.1016/j.immuni.2018.03.023.
2. Auslander N, Zhang G, Lee JS, Frederick DT, Miao B, Moll T, Tian T, Wei Z, Madan S, Sullivan RJ, *et al*. Robust Prediction of Response to Immune Checkpoint Blockade Therapy in Metastatic Melanoma. *Nat. Med* 2018; **24**(10): 1545. https://doi.org/10.1038/s41591-018-0157-9.

Combination immunotherapies

P855 HIGH-RESOLUTION MAPS OF HETEROGENEOUS ANTIGEN EXPRESSION IN GLIOBLASTOMA AND IMPLICATIONS FOR IMMUNOTHERAPY

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10.1136/LBA2019.9

Background Glioblastoma (GBM) remains an almost universally fatal brain tumor. While CAR T cell immunotherapy has shown promising clinical efficacy, therapeutic failure may reflect our incomplete understanding of target antigen expression. We previously examined variations in antigen expression at the level of individual patients (inter-patient or inter-tumor heterogeneity), focusing on immunotherapy targets IL13Rα2, HER2 and EGFR. We concluded that antigen expression diverged from expectations from random expression. Because antigen escape may arise from GBM cell heterogeneity, we have mapped target antigen expression within individual tumors (intra-tumor heterogeneity).

Methods Serial sections from a 43 patient cohort were immunostained (DAB with hematoxylin counterstain) for target antigens IL13Rα2, HER2 and EGFR. Each section was annotated directly from the slide by a neuropathologist. Sections were scanned (0.46 μm/pixel; Hamamatsu), and then working within Fiji/ImageJ, images were segmented by color deconvolution into hematoxylin (nuclei) and DAB layers. Images of nuclear layers were aligned, and used to align the DAB layers. Two schemes were used to examine the spatial distributions of the three target antigens. When tumor domains could be identified, we determined expression of each antigen as optical density (OD). In the second scheme, a 10 μm grid was superimposed on each section, and OD was determined for each position and assembled into spreadsheets (Origin v2019b). Maps for expression were generated from the OD in each position.

Results Approaching these maps from the perspective of antigen escape, we examined the extent to which expression of target antigens was spatially mixed, how rapidly antigen dominance could shift (spatial frequency), and whether spatial distributions were arrayed in a coordinated manner.

When tumor domains could be identified, we calculated the Shannon diversity index (H) for each domain within a section. While values of H clustered within some tumors, usually values of H varied widely.

The superimposed grid was used to examine heterogeneity within entire tumor sections. Expression was intermixed, and EGFR and IL13Rα2/HER2 displayeds complementary expression patterns. In tumors with large EGFR+ areas, IL13Rα2+/HER2+ areas could overlap, while when EGFR+ areas were smaller, IL13Rα2+ and HER2+ areas were more distinct. Borders could be quite diffuse, or quite sharp (a few cell diameters).

Conclusions Our results indicate that expression of IL13Rα2, HER2 and EGFR is highly heterogeneous and not always spatially distinct. Because GBM tumors adapt to the selection pressures of immunotherapies, we suggest that combination therapies should be designed accordingly, and immunotherapies targeting IL13Rα2/HER2 could benefit from inclusion of EGFR.

Completed clinical trial

P856 AVID200, FIRST-IN-CLASS TGF-BETA1 AND BETA3 SELECTIVE INHIBITOR: RESULTS OF A PHASE 1 MONOTHERAPY DOSE ESCALATION STUDY IN SOLID TUMORS AND EVIDENCE OF TARGET ENGAGEMENT IN PATIENTS

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10.1136/LBA2019.10

Background AVID200 is a rationally designed first-in-class receptor ectodomain trap that inhibits transforming growth factor-beta (TGF-beta) isoforms -beta1 and -beta3 with pM