Simultaneous Characterization and Quantification of Immune Cell Subpopulations and PD-L1 Expressing CTCs in Peripheral Blood of Cancer Patients

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Background

- Expression of PD-L1 on tumor, and immune markers in tumor tissue, are associated with improved response to PD-1 and PD-L1 checkpoint inhibitors. However, each alone has limited predictive utility.
- Multimodal characterization of both the tumor and host immune system is an unmet medical need for the improved prediction of response to immunotherapy.
- Metastatic lesions are likely to be under-sampled and require a liquid biopsy, given tumor heterogeneity and evolution and temporal changes in the host immune system.

- We sought to examine expression of PD-L1 on CTCs as well as characterize rare immune cell populations with a non-invasive liquid biopsy. Examining dynamic biomarker changes in longitudinal samples could enable the development of novel diagnostic tools for response prediction and pharmacodynamics studies related to immunotherapy.

Methods

Blood samples were drawn from 3 healthy donors, 5 non-cancerous lung disease patients, and 4 metastatic cancer patients (prior to and on therapy with checkpoint inhibitor), and sent to Epic Sciences for processing with Epic Sciences’ CTC and immune cell assays.

1) SLIDE PREPARATION
2) CELL STAINING
3) SCANNING
4) SINGLE CELL DIGITAL PATHOLOGY

PD-L1(+) Peripheral Circulating Cells in Lung Cancer

Tools to Characterize Rare Immune Cell Populations

Schematic of Epic’s CTC platform: CTC enumeration, leukocyte classification, morphology, and protein analysis

- Nucleated cells from 8-10 mL blood were enriched, sorted, and analyzed using a single sample per slide (approximately 1 million total).
- Slides are scanned using a rapid fluorescent scanning method, which images each nucleated cell.
- All cells are analyzed with a multi-parametric digital pathology algorithm for morphological features and protein expression.

Conclusions

- Multimodal characterization of both the tumor and host immune system is an unmet medical need for the improved prediction of response to immunotherapy.
- Detection of PD-L1 in circulating cells from NSCLC liquid biopsy samples was associated with worse overall survival, highlighting the potential utility of minimally invasive liquid biopsies for tumor profiling.
- Epic Sciences has developed tools to simultaneously detect, characterize, and quantify CTCs and rare immune cell subpopulations from a single blood draw.
- Examining dynamic biomarker changes with minimally invasive liquid biopsies in longitudinal samples could enable the development of novel diagnostic tools for response prediction and pharmacodynamics studies related to immunotherapy.

Changes in CD4/CD8/Ki-67 with PD-L1 inhibitor Tx

Changes in CD4, CD8, and Ki-67 positive populations were detected in metastatic cancer patient samples upon treatment with PD-L1 inhibitor. Changes in CD4, CD8, and Ki-67 were measured in blood of lung cancer patients and is associated with worse survival. PD-L1 expression was also measured in a matched cohort of healthy donors.

Cell Ratios (CD4/CD8)

- Percent CD4+ (Target)
- Percent CD8+ (Target)

Changes in CD4/CD8/Ki-67 in Lung Disease Patients

Schematic of Epic Single Cell Tools for Immune Monitoring

- Nucleated cells from 8-10 mL blood were enriched, sorted, and analyzed using a single sample per slide.
- Slides can be used for CTC and immune cell characterization as well as downstream genomic analysis.

Changes in CD4+ and CD8+ cell populations were detected in lung cancer patients upon treatment with PD-L1 inhibitor. Changes in CD4, CD8, and Ki-67 were measured in blood of lung cancer patients and is associated with worse survival. PD-L1 expression was also measured in a matched cohort of healthy donors.

Graphs show changes in CD4 and CD8 positive populations. A) Percent change in CD4+ and CD8+ cells in healthy donors compared to lung cancer patients. B) Percent change in CD4+ and CD8+ cells in healthy donors compared to lung cancer patients. C) Percent change in CD4+ and CD8+ cells in healthy donors compared to lung cancer patients. D) Percent change in CD4+ and CD8+ cells in healthy donors compared to lung cancer patients.