64 CANCER TESTIS ANTIGEN CO-EXPRESSION LANDSCAPE IN SOLID TUMORS

RJ Seager*, Erik Van Roey, Shuang Gao, Jonathan Andreas, Vincent Giamo, Blake Burgher, Felicia Lenzo, Mary Nesline, Paul DePietro, Yirong Wang, Sean Glenn, Shengle Zhang, Jeffrey Conroy, Sarabjot Pabla. *OmnSeq, Buffalo, NY, USA*

Background Cancer testis antigens (CTAs) are tumor antigens that have a highly tissue-restricted expression but are often expressed in diverse malignancies. With their highly immunogenic expression limited to tumor cells, CTAs have become a prime target for cancer vaccinations and T-cell-based therapy with chimeric T-cell receptors. In this study, we investigated the landscape of 17 CTA (NY-ESO-1, LAGE-1A, and 15 other CTAs) in the context of the tumor immune microenvironment of real-world clinical tumors spanning multiple histologies.

Methods RNA-seq was performed on 5450 FFPE tumors, and the expression of each of the 17 CTAs were classified as Positive (nRPM≥20) or Negative (nRPM<20). Pearson correlation analysis was conducted on the nRPM values for each CTA to determine co-expression relationships between any of the 17 CTAs. In order to visualize patterns in the CTA expression landscape, heatmap analysis was performed, using hierarchical clustering with Pearson's correlation as a distance measure to reveal patterns in CTA status across all samples and CTAs.

Results 5450 tumor samples analyzed in this study spanned 39 histologic types of tumor and were predominantly composed of lung cancer (40.4%) followed by colorectal cancer (10.6%) and breast cancer (8.6%). Positive CTA prevalence ranged from 2.4% (GAGE13) to 31.5% (XAGE1B). A high degree of significant correlation between the expression of all CTAs was observed, with only GAGE10, XAGE1B, MLANA, MAGEA4, GAGE13, and SSX2 having a no significant correlation with at least one other CTA. Three key groups of co-expressed CTAs were observed: 1) NY-ESO-1, LAGE-1A, MAGEA12, MAGEA3, MAGEA1, MAGEA10, and MAGEA4 (0.36≤R≤0.82); 2) GAGE12J, GAGE2, GAGE1, GAGE13 (0.58≤R≤0.72); 3) SSX2, BAGE, MAGEC2 (0.4≤R≤0.58). The three remaining CTAs (GAGE10, XAGE1B, and MLANA) had little or no (R<0.22) correlation with any other CTA or each other. Clustering CTAs across all samples revealed three CTA expression clusters: 1) samples that express a collection of multiple CTAs; 2) samples that express mostly XAGE1B, over-represented by lung cancer (p=1.51e-296); 3) samples that express mostly GAGE10, over-represented by neuroendocrine tumors (p=1.64e-05).

Conclusions Across multiple cancer subtypes, the expression of a CTA occurs in the context of other CTAs, and specific groups of CTAs are likely to co-express, forming expression patterns characteristic to tumor subgroups. These findings provide a scientific base for selecting appropriate CTAs and designing multiplex vaccination in immunotherapies of a variety of tumors. Further studies are needed to understand the relationship of these CTAs with traditional and emerging immune-oncology biomarkers.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0064

65 PD-L1 BY RNA NEXT GENERATION SEQUENCING: COMPARISON WITH PD-L1 IHC 22C3 AND ASSOCIATION WITH SURVIVAL BENEFIT FROM PEMBROLIZUMAB WITH OR WITHOUT CHEMOTHERAPY IN NON-SMALL CELL LUNG CANCER

[1]Yong Hee Lee*, [2]Grace Dy, [1]Paul DePietro, [1]Jeffrey Conroy, [1]Sarabjot Pabla, [1]Mary Nesline. *[1]OmniSeq, Buffalo, NY, USA; [2]Roswell Park Comprehensive Cancer Center, Buffalo, NY, USA*

Background PD-L1 immunohistochemistry (IHC) testing is suboptimal for predicting patient clinical benefit for checkpoint inhibition, while PD-L1 liquid biopsy is not clinically validated and lacks sensitivity, underscoring the need to include PD-L1 testing in more robust, tissue-efficient, comprehensive, scalable next generation sequencing (NGS) tests.

Methods To assess comparability and efficacy of PD-L1 testing by NGS with IHC, we identified NSCLC patients treated by first-line pembrolizumab alone (n=54) or pembrolizumab + chemotherapy (n=49) whose tumors underwent companion diagnostic PD-L1 testing by IHC antibody 22C3 testing (high≥50%; low=1–49%, or negative=0% tissue proportion score), and also by RNA-seq, as part of a comprehensive immune profiling panel. PD-L1 expression by RNA-seq, was measured as a percentile rank, with ≥75 considered 'high', and <75 considered 'not high', based on comparison to a reference population and normalized to a value of 1–100. All testing was performed in a CLIA certified laboratory prior to treatment initiation (any line) at Roswell Park Comprehensive Cancer Center (June 2017-March 2019, with a minimum of 1 year of follow up). Assay equivalence was assessed by proportion analysis using Fisher exact test comparing IHC versus to RNA-seq, and Bonferroni pairwise post-hoc analysis of IHC (high vs. low, high vs. negative, low vs. negative) with RNA-seq (high vs. not high). A Cox regression model evaluated associations between IHC and RNA-Seq with OS from first dose of pembrolizumab.

Results More than 75% of IHC high cases were classified as high by RNA-Seq for both treatment groups (p<0.001). Post-hoc pairwise comparisons showed PD-L1 IHC and RNA-Seq 'high' results were significantly associated with each other, and PD-L1 IHC low/negative results were associated with RNA-seq 'not high' results. In the pembrolizumab monotherapy group, RNA-seq high was associated with improved survival for pembrolizumab compared to RNA-seq not high status (HR=3.96; CI=1.22–12.87; p=0.02), while PD-L1 IHC high status was not associated with survival benefit in this group (p=0.63). In the pembrolizumab + chemotherapy group, as expected, neither IHC (high versus low), nor RNA-seq (high versus not high) status was associated with survival benefit (p=0.81 and p=0.76, respectively). These findings are consistent with our previous work demonstrating PD-L1 RNA-seq was predictive of CPI response in multiple tumor types.

Conclusions PD-L1 status by RNA-seq and IHC appear to be comparable. Unlike PD-L1 IHC however, PD-L1 RNA-seq high status versus not high status is associated with greater survival benefit, indicating PD-L1 by NGS may have utility for pembrolizumab selection.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0065