OTR.22 - Immune landscape in esophageal squamous cell carcinoma

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Introduction:

Esophageal cancer (EC) is one of the ten most incident and lethal neoplasms worldwide. The chemotherapy of choice still involves taxane and platinum-based regimens, without any molecular targets. Therefore, it is of utmost importance to better characterize these tumors in order to develop biomarkers and new therapeutical strategies. Esophageal squamous cell carcinoma (ESCC) exhibits high intratumoral molecular heterogeneity that might favor immunotherapy, such as the immune checkpoints blockade. Nonetheless, the success of such therapies depends on the immune based microenvironment characteristics of the tumor.

Objective:

Describe the immune cell infiltrate in ESCC tumor, find prognosis biomarkers and propose new treatment strategies to ESCC patients.

Methodology:

RNA-seq from 14 tumor and adjacent normal tissue samples from ESCC patients without previous treatment (from BNT, INCA - CEP 116/11) were performed by Illumina Hi-Seq 2000. Mutations were analyzed by GATK best protocols. Neoantigens were predicted based on NetMHCpan 3.30 and HLA alleles by Optitype. CIBERSORT was used to calculate 22 immune cell subpopulations percentage. MiXCR and tcR package was applied to estimate TCR and BCR clonotypes. R packages were used for graphs and statistics.

Results:

We found 75 mutations per sample, resulting in 29 neoantigens in average. Tissue associated antigens (TAA) expression was evaluated and five genes were specifically expressed in tumor samples, but not in surrounding healthy tissue. Peptides derived from these genes could be used as a vaccine. Immune cell infiltrate populations were evaluated resulting in a major population of macrophages and T cell within the tumor. Cytokines and chemokines gene expression also correlated with several immune subpopulations. B memory cells correlates with better prognosis both in INCA and TCGA cohorts. BCR repertoire were predominantly IgG whereas IgA was in surrounding area. We also found germinative centers within the tumor by immunohistochemistry staining. TCR and BCR number of clonotypes correlated with neoantigen burden. TCR alpha and beta chains were the most abundant. Several clonotypes of TCR and BCR were shared between tumor and surrounding tissue, with very few shared between samples. By *in silico* molecular model we found a TCR clone that recognized a TAA derived peptide from MAGEA11. Concerning immune checkpoint blockade molecules, we tested several inhibitors and stimulatory receptors as *lag3, pdcd1, pdl1* and *ctl4* that were more expressed in tumor and higher *lag3* expression correlated with a poor prognosis in INCA cohort.

Conclusion:

We described the complex role of several immune cell subsets, its inhibitory and stimulatory molecules in mucosa collaborating for the tumor microenvironment. Also, we found a TCR specific for a neoantigen. Altogether these results showed that each patient has its own collection of neoantigens derived from mutations, but shared peptides from TAA genes that can be proposed as a unique peptide vaccine for ESCC patients.

Keywords: Immune system; Esophageal Cancer; Neoantigen