REDUCED DENDRITIC CELL INFILTRATION IN ANTI-PD-1 RESISTANT HEAD AND NECK SQUAMOUS CELL CARCINOMA

1Shin Saito*, 1Hirofumi Shibata, 1Liye Zhou, 3Katie Campbell, 3Ann Marie Egloff, 1Ravindra Uppaluri.
1Dana Farber Cancer Institute, Boston, MA, United States; 2University of California Los Angeles, Los Angeles, CA, United States; 3Brigham and Women’s Hospital, Boston, MA, United States

Background Although immune checkpoint inhibitors have been approved for head and neck squamous cell carcinoma (HNSCC) patients, the majority do not respond, and further treatment optimization is required. Diverse anti-PD-1 resistance mechanisms have been proposed for this lack of benefit, which is inconsistent with the moderate tumor mutational burden and immune infiltration observed in HNSCC. Here we identified neoantigen expression and characterized dendritic cell (DC) infiltration in previously reported murine oral carcinoma (MOC) models with differential response to anti-PD-1, and we interrogated DC infiltration as a correlate of response in a neoadjuvant anti-PD-1 clinical trial.

Methods HNSCC MOC1 and its isogenic anti-PD-1 resistant MOC1-esc1 models were subjected to immunogenomic analysis. MOC1 and MOC1-esc1 expressed neoantigens that were defined using whole exome and RNA sequencing (RNASeq) data from cultured lines, and Class I presented neoantigens were predicted in silico (NetMHCpan4.0). MOC1 tumor infiltrating lymphocytes (TILs), following control, anti-PD-1 or anti-CTLA-4 treatment, were assayed for reactivity to synthesized predicted neoantigen peptides, and DC tumor infiltration was assessed by flow cytometry. Patient HNSCC tumor RNA-Seq data from our neoadjuvant pembrolizumab trial (NCT02296684) were interrogated, and response to neoadjuvant anti-PD-1 was classified based on pathological response in surgical specimens.

Results As we previously reported1, MOC1 was anti-PD-1 responsive and MOC1-esc1 was resistant, while both lines were sensitive to anti-CTLA-4 therapy. Results from our immunogenomics pipeline identified 325 neoantigen candidates in MOC1 with a predicted Class I binding affinity (% rank) of less than 0.5. We tested 35 putative neoantigens for TIL immune reactivity to synthesized candidate neoepitope peptides using ELISPOT. MOC1 TIL had reactivity to mutant Yipf1 (mYipf1) neoepitope in addition to the endogenous retrovirus derived p15E antigen. Anti-PD-1 modestly increased while anti-CTLA-4 significantly increased MOC1 TIL reactivity to p15E and mYipf1 compared to control. Analysis of RNA-Seq data from MOC1 and MOC1esc1 showed a reduced conventional DC1 (cDC1) signature in MOC1esc1, and we confirmed this reduced infiltration by flow cytometry. Finally, in neoadjuvant anti-PD-1 HNSCC trial samples, a published DC signature [2] was found to be significantly higher in responder pretreatment biopsies compared to non-responders (p=0.016).

Conclusions We identified mYipf1 as a novel neoantigen in MOC1 and employed TIL response to mYipf1 or p15E peptides to measure MOC TIL anti-tumor activity. CTLA-4 directed antibodies enhanced TIL anti-tumor activity compared to anti-PD1 treatment. Reduced cDC1 infiltration and lack of neoantigen expression may represent an anti-PD-1 resistance mechanism in HNSCC.

REFERENCES

Ethics Approval All patients provided informed consent prior to specimen donation.

Patient samples were analyzed under the auspices of Dana Farber Cancer Institute IRB approved protocol 18-092 (PI Uppaluri).
Animal studies were performed under Dana Farber Cancer Institute IACUC protocol 16-020 (PI Uppaluri).