

Neoantigens

Major finding: Neoantigens derived from oncogenic gene fusions can elicit T-cell responses in tumors with low TMB.

Concept: Gene fusions represent a source of tumor-specific neoantigens.

Impact: Oncogenic gene fusions expressed in tumors may have therapeutic potential for immunotherapy.

ONCOGENIC GENE FUSION-DERIVED NEOANTIGENS ELICIT SPECIFIC T-CELL RESPONSES

Antitumor immunity is mediated by the recognition of tumor-specific neoantigens by cytotoxic T cells. For this reason, immune checkpoint inhibitor therapies are generally more effective in cancers with high tumor mutational burden (TMB). Gene fusions are oncogenic drivers in many cancers with low TMB but have not been identified thus far as a source of neoantigens despite the likelihood that they would produce neoantigens more immunogenic than those derived from missense mutations. Yang, Lee, Srivastava and colleagues identified a novel *DEK-AFF2* gene fusion in a patient with head and neck squamous cell carcinoma whose tumor had low TMB and minimal immune infiltrate but a complete response to the anti-PD-1 antibody pembrolizumab. Analysis of predicted HLA-restricted peptides revealed a *DEK-AFF2*-derived peptide that stimulated the activation of T cells from the patient. The fusion peptide was responsible for the response to anti-PD-1 treatment and tumor regression. To study gene fusion-derived neoantigens in other cancers with low TMB, a panel of adenoid cystic carcinoma head and neck cancers expressing *MYB-NFIB* fusions was screened for HLA-A2-restricted peptides. The screen uncovered three



peptides derived from the *MYB-NFIB* fusion and one peptide derived from an *NFIB-MYB* fusion. T cells from peripheral blood mononuclear cells of a patient expressing the *MYB-NFIB* fusion showed reactivity to the fusion peptide. The presence of fusion-derived peptides was further analyzed in 30 cancer types in The Cancer Genome Atlas. Fusion-derived HLA-predicted peptides were found in 24%

of all fusion-expressing cancers and were most likely to be detected in the presence of low immune surveillance. In addition, analysis of a cohort of tumors from patients with melanoma treated with anti-PD-1 therapy revealed additional predicted fusion neoantigens that were observed to be eliminated during successful treatment. Collectively, these results propose gene fusions that are expressed in low TMB cancers as a source of immunogenic neoantigens that can elicit T-cell responses and potentially have important implications for designing immunotherapeutic approaches. ■

Yang W, Lee KW, Srivastava RM, Kuo F, Krishna C, Chowell D, et al. Immunogenic neoantigens derived from gene fusions stimulate T cell responses. *Nat Med* 2019;25:767–75.

Immunotherapy

Major finding: Tumor-initiating stem cells exhibit enhanced resistance to immunotherapy via expression of CD80.

Concept: CD80 expression on tumor-initiating stem cells dampens cytotoxic T-cell antitumor activity.

Impact: Inhibition of CTLA4 or TGFβ are potential strategies to restore stem-cell sensitivity to immunotherapy.

TUMOR-INITIATING STEM CELLS ARE RESISTANT TO IMMUNOTHERAPY

Tumor-initiating stem cells (tSC) possess the ability to self-renew and differentiate at low numbers, granting them the capacity for tumor growth and relapse following therapy. Despite their suspected role in resistance to immunotherapies such as adoptive cell transfer (ACT), the precise mechanisms by which tSCs escape immune surveillance remain unknown. Miao and colleagues utilized an immunotherapy-responsive mouse model of squamous cell carcinoma (SCC) to show that tSCs evade immune detection and initiate tumor relapse following ACT. Injection of tumor-specific cytotoxic T lymphocytes (CTL) resulted in robust infiltration into the tumor and significant reduction of tumor volume. Residual tumor cells persisted beyond the point of CTL exhaustion and survived a second treatment of ACT. Single-cell RNA sequencing of residual tumor cells identified a distinct cluster of tSCs expressing TGFβ with reduced expression of proinflammatory cytokines, chemokines, and proapoptotic genes. These tSCs also expressed CD80, a surface ligand that binds to the

immune checkpoint protein CTLA4. Treatment with a TGFβ-blocking antibody reduced surface CD80 expression in tSCs, and treatment with a CTLA4-blocking antibody reinvigorated CTL activity against CD80⁺ SCC cells *in vitro* and blunted SCC tumor growth *in vivo*. Genetic ablation of *Cd80* *in vivo* impaired tumor growth and enhanced CTL infiltration, proliferation, and antitumor activity, and specific deletion of *Cd80* or *Ctla4* in TGFβ-expressing tSCs resulted in more frequent and more active CTLs with diminished expression of inhibitory receptors, resulting in increased apoptosis in tSCs, decreased tSC survival, and diminished tumor relapse following ACT. Taken together, these findings establish a role for CD80 in tSC resistance to immunotherapy and highlight several potential therapeutic strategies to overcome this resistance. ■

Miao Y, Yang H, Levors J, Yuan S, Polak L, Sribour M, et al. Adaptive immune resistance emerges from tumor-initiating stem cells. *Cell* 2019;177:1172–86.