Prostate Cancer

Major Finding: Enzalutamide-treated prostate cancer and small-cell carcinoma cells had distinct transcriptomes.

Concept: Enzalutamide is an androgen-targeting drug, and small-cell carcinoma resists androgen-targeted drugs.

Impact: This study characterizes the transcriptomic impact of androgen-targeted treatment in prostate cancer.

SINGLE-CELL TRANSCRIPTOMICS REVEAL TREATMENT EFFECTS IN PROSTATE CANCER

In metastatic castration-resistant prostate cancer (mCRPC), intrinsic or acquired resistance to second-generation androgen-targeting therapies including enzalutamide and abiraterone is the norm. Although several studies have implicated individual mutations and dysfunctions in certain pathways as drivers of resistance, considerable knowledge gaps still exist. To address this challenge, He and colleagues conducted single-cell transcriptomic analyses on tumor samples from 14 patients; these samples comprised 15 fresh biopsies from mCRPC tumors representing common sites of metastasis (bone, lymph node, and liver). Most profiled tumors were adenocarcinomas, but one had small-cell carcinoma histology. Despite the fact that mCRPCs are commonly characterized by expression of various androgen receptor (AR) isoforms—many of which lack the ligand binding domain and are hypothesized to be constitutively active—no association between any specific isoform and prior enzalutamide exposure was found. Instead, mCRPC cells ubiquitously expressed mRNAs encoding alternate AR isoforms regardless of prior treatment. However, cells from enzalutamide-treated tumors did exhibit some differences: For example, these cells had increased expression of gene sets related to the epithelial–mesenchymal transition and TGFβ signaling, and the latter finding was supported by *in vitro* experiments. Notably, cancer cells from the biopsy of small-cell carcinoma—a subtype of neuroendocrine prostate cancer (NEPC) that is resistant to androgen-targeted therapies—had no detectable *AR* transcripts and had downregulation of an AR-regulated gene set and upregulation of an NEPC gene set. Further, cells from the small-cell carcinoma biopsy had higher expression of *NANOG, SOX2*, and *EZH2*, all of which have been associated with lineage plasticity and resistance to androgen-targeting therapies, along with the transcription factor–encoding genes *HOXB5, HOXB6*, and *NR1D2*. Finally, in one patient, tumor-infiltrating cytotoxic CD8$^+$ T cells that were undergoing clonal expansion following enzalutamide treatment were characterized by elevated expression of dysfunction-associated markers, including *PDCD1* (encoding PD-1). Together, these findings provide a detailed characterization of the transcriptional states of cancer cells and infiltrating immune cells in the mCRPC microenvironment and provide insight into the impacts of androgen-targeted treatment. ■

He MX, Cuoco MS, Crowdis J, Bosma-Moody A, Zhang Z, Bi K, et al. Transcriptional mediators of treatment resistance in lethal prostate cancer. Nat Med 2021;27:26–33.

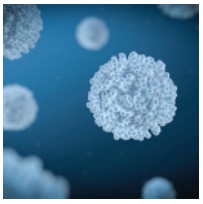
Immunology

Major Finding: An *in vivo* CRISPR screen found that the transcription factor FLI1 inhibited effector CD8$^+$ T cells.

Concept: FLI1 depletion increased RUNX3 chromatin accessibility, enhancing effector T-cell differentiation.

Impact: This work reveals FLI1 as a novel target to derepress effector T cells in cancer and infection.

IN VIVO CRISPR SCREEN IDENTIFIES FLI1 AS REGULATOR OF EFFECTOR T CELLS



Immunotherapy aims to enhance antitumor immune responses, but the duration of patient benefit is often limited, in part due to T-cell exhaustion. To investigate pathways regulating differentiation into exhausted (T_{EX}) versus effector (T_{EFF}) CD8$^+$ T cells, Chen and colleagues developed OpTICS, an *in vivo* CRISPR screening approach in which Cas9$^+$ antigen-specific murine CD8$^+$ T cells are transduced with a library of single guide RNAs targeting 120 transcription factor genes. T cells were then adoptively transferred into recipient mice, and the OpTICS platform identified candidates that inhibited optimal T-cell activation and T_{EFF}-cell differentiation in models of lymphocytic choriomeningitis virus (LCMV) infection. The gene encoding the ETS family transcription factor FLI1 was a top hit that repressed T_{EFF} cell differentiation, and further investigation revealed that CRISPR–Cas9-mediated mutagenesis of *Fli1* increased CD8$^+$ T-cell proliferation and T_{EFF}-cell populations during acutely resolved LCMV infection. Similarly, during chronic LCMV infection, FLI1 depletion in antigen-specific CD8$^+$ T cells enriched for a signature of T_{EFF}-cell gene expression, supporting the idea that FLI1 deficiency promoted T_{EFF}-cell fate. Mechanistically, loss of FLI1 in CD8$^+$ T cells shifted chromatin accessibility, especially in intronic or intergenic regions near known T_{EFF}-cell genes. ETS and RUNX motifs were highly enriched in regions with increased accessibility in the absence of FLI1. Given that the RUNX family member RUNX3 has been well established as a central driver of T_{EFF}-cell differentiation, RUNX3 was hypothesized to promote T_{EFF}-cell fate downstream of FLI1 loss. Indeed, overexpression of RUNX3 in the absence of FLI1 enhanced the CD8$^+$ T-cell response to LCMV infection and skewed the T-cell population toward T_{EFF}-like cells. Importantly, loss of FLI1 in CD8$^+$ T cells not only improved protective immunity in several models of viral or bacterial infection but also enhanced antitumor immunity, controlling tumor progression in a subcutaneous B16 melanoma model. Together, these results show that FLI1 inhibits T_{EFF}-cell fate by restricting RUNX3 genome access, highlighting FLI1 as a potential immunotherapy target that may enhance optimal T_{EFF}-cell differentiation in cancer and infection. ■

Chen Z, Arai E, Khan O, Zhang Z, Ngiow SF, He Y, et al. In vivo CD8$^+$ T cell CRISPR screening reveals control by Fli1 in infection and cancer. Cell. 2021;184:1262–80.