

TCR functional promiscuity and cytotoxic CD8 T cell expansion drive myocarditis induced by immune checkpoint inhibitors



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ABSTRACT

Introduction

Immune checkpoint inhibitor-associated myocarditis is a serious immune-related adverse event (irAE) that can threaten life and often results in stopping immunotherapy. This complication makes patient management more difficult, especially in cases needing high-dose steroids, which may later encourage tumor growth. To enhance understanding of disease mechanisms, we examine T-cell clonal populations in the peripheral blood of patients with ICI-related cardiotoxicity.

Methods

We performed single-cell RNA sequencing, coupled with paired TCR sequencing, of circulating T cells from ICI-associated myocarditis patients before and after treatment with Atezolizumab. A computational approach was applied to perform clonal, transcriptional, and biophysical analyses, integrated to characterize disease-associated CD8 T cell responses, with a focus on clonally expanding effector populations and the structural properties of their CDR3 hypervariable regions.

Results

Patients with ICI-associated myocarditis exhibited differences in cellular and TCR repertoires. Effector CD8 T cells were significantly elevated at baseline and after ICI treatment compared with patients without irAE. This finding was confirmed in the pericardial fluid and in a dataset of T cells obtained from human heart tissue affected by ICI-myocarditis patients. The TCR sequencing results notably showed that clonally expanded CD8 T cells expressing PRF1 and GZMK also had increased GZMA expression, along with higher levels of activation markers CD69 and KLRG1. These data indicate that a distinct clonally expanded subpopulation within the CD69-expressing effector CD8 T cell compartment maintains a pathogenic, pro-inflammatory state in ICI-myocarditis patients. The expanded T cell clones displayed shorter TCR-CDR3 sequences and a significantly higher proportion of hydrophobic residues within the CDR3 region.

Conclusion

Our findings highlight the role of specific effector CD8 T cell subsets in promoting cardiac inflammation. TCR physical properties, for example, length and hydrophobicity, provide a novel mechanical insight into T cell-associated irAE pathogenesis. These results establish a foundation for leveraging TCR biophysical characteristics to selectively target pathogenic T cells, thereby mitigating myocarditis risks and enabling more precise therapeutic interventions for vulnerable patients.

SCIENTIFIC IMPACT

This study investigates the mechanisms underlying immune checkpoint inhibitor (ICI)-associated myocarditis, with its primary contribution being the identification of biophysical TCR constraints associated with this irAE. Pathogenic clones were characterized by shorter CDR3 sequences and significantly increased hydrophobicity within the CDR3 region. These structural features suggest a shift toward lower-affinity or highly cross-reactive T-cell receptor interactions, potentially facilitating the breakdown of cardiac self-tolerance. By moving beyond simple clonal expansion metrics, these findings introduce a novel biophysical framework for identifying "at-risk" TCR repertoires and informing the development of targeted, steroid-sparing interventions to mitigate fatal cardiotoxicity without compromising antitumor efficacy.