CELL-INTRINSIC TIM-3 IS REQUIRED FOR OPTIMAL CD8 RESPONSE TO ACUTE LCMV INFECTION

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Background Tim-3, or transmembrane immunoglobulin and mucin domain-3, is a type I membrane protein expressed by various immune cell types and which has been shown to have a co-stimulatory role in T cells through the PI3K pathway. Comprising an extracellular, a transmembrane and an intracellular domain containing five tyrosine residues, we and others have shown that Tim-3 participates in TCR-mediated signaling pathways. Using LCMV infection, we also found that Tim-3 expression influences the formation of short-lived effector and memory precursor CD8+ T cells. We hypothesize that Tim-3 enhances CD8+ T cell function in acute infections and contributes to enhanced CD8+ effector function.

Methods To test this hypothesis, we have used LCMV-specific TCR transgenic (P14) mice expressing a truncated version of Tim-3 and CD8+ conditional deletion of Tim-3. CD8+ T cell activation and memory were analyzed by flow cytometry after acute infection with LCMV Armstrong. Bulk RNA and TCR sequencing was performed on endogenous Tim-3 expressing and non expressing effector CD8+ cells. Nuclear localization of Foxo1 was analyzed using Amnis Imagestream.

Results At the effector stage, Tim-3+ T cells had increased expression of effector CD8 markers such as T-bet. Tim-3 knockout mice also showed a significantly lower number of LCMV-specific T cells. Additionally, endogenous Tim-3+ T cells were also significantly better at cytokine production and had increased cytotoxicity. Tim-3 expression correlated with better cell survival in homeostatic conditions and during reactivation induced cell death. Through bulk RNA and TCR sequencing, we have found that Tim-3+ effector CD8+ T cells also have a distinct pattern of upregulation of effector genes and show increased expansion of TCR clones. Mechanistically, we also show that Tim-3 signals via Foxo1, inhibiting its entry into the cytoplasm, thus conferring an enhanced effector function.

Conclusions Our data suggest that Tim-3 signaling positively contributes to an effective CD8+ T cell response in an acute infection. These findings may lead to a better understanding of CD8+ T cell function in different settings, including in the tumor microenvironment.

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REFERENCES
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Ethics Approval All mouse experiments were performed in accordance with protocols approved by the University of Pittsburgh Institutional Animal Care and Use Committee

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