EXHAUSTED T CELLS ARE CHARACTERIZED BY INAPPROPRIATE LIPID ACCUMULATION

Kellie Spahr*, Nicole Scharping, Greg Delgoffe. University of Pittsburgh, Pittsburgh, PA, United States; University of California – San Diego, San Diego, CA, United States

Background The efficacy of immunotherapy depends on the presence and persistence of functional immune cells within the tumor. While tumor-specific T cells can be activated and infiltrate the tumor microenvironment, they are quickly rendered dysfunctional by the combination of chronic antigen stimulation, hypoxia, and nutrient stress that creates a uniquely immune-suppressive environment. Thus, T cell dysfunction remains a significant hurdle for immunotherapeutic success. We have shown T cell exhaustion and metabolic dysfunction are driven by mitochondrial stress. These features evoke an image of weak, starving T cells that are unable to sufficiently fuel their effector function. However, we and others have observed that CD8+ T cells accumulate large lipid stores as they progress towards exhaustion. What remains unclear is whether lipid accumulation in these cells contributes to their dysfunction or represents an untapped source of carbon that may be the key to their reinvigoration.

Methods Using an in vitro model of T cell exhaustion developed in our lab, we evaluated the effect of perturbing lipid metabolism by inhibiting synthesis or promoting catabolism. We also used CRISPR-Cas9 deletion of the mitochondrial citrate transporter, SLC25A1, to assess the effect of perturbing lipid synthesis on tumor-infiltrating T cell exhaustion. Cogenically mismatched SLC25A1 deficient and control OT-I T cells were co-transferred into mice bearing ovalbumin-expressing B16F10 melanoma tumors. We immunophenotyped CD8+ T cells for markers of terminal exhaustion, cytokine production, and lipid accumulation.

Results Here we sought to define the role lipid metabolism plays in the progression of CD8+ T cell exhaustion. Inhibition of citrate transport from the mitochondria via SLC25A1 in CD8+ T cells resulted in reduced lipid accumulation and expression of inhibitory receptors, PD-1 and Tim-3, known to be upregulated on exhausted T cells. We also observed increased production of inflammatory cytokines IFNγ and TNF in response to TCR restimulation after chronic antigen exposure.

Conclusions Taken together, our results indicate a role for mitochondrial citrate flux in the accumulation of cytosolic lipids and progression of CD8+ T cell exhaustion. We propose that as exhausted T cells experience mitochondrial stress and perform less oxidative phosphorylation, they shuttle excess citrate to the cytosol where it fuels production of acetyl-CoA and de novo fatty acid synthesis. This pathway may be targeted to delay exhaustion or reinvigorate exhausted T cells within tumors. These data provide new insight into the mechanisms of T cell exhaustion and may inform future immunotherapeutic development.

REFERENCES