A LIVE TUMOR FRAGMENT PLATFORM SUITABLE FOR ASSESSING RESPONSE TO T-CELL THERAPIES

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Background T-cell therapies have shown efficacy in numerous clinical settings. However, predictive biomarkers of response are lacking, and determining which T-cell therapy each cancer patient will respond to has been hindered by the absence of model systems that maintain the specific immunosuppressive features of each patient’s tumor microenvironment (TME). The Elephas live tumor fragment (LTF) platform is designed to enable prediction of clinical response to many types of immunotherapies, including cellular therapies and checkpoint blockade. The experiments described here examine T-cell activation, cytokine production, and tumor-cell killing following co-culture of LTFs with T-cell therapies.

Methods Transgenic T-cells: LTFs were generated from two tumors that differ only by the presence or absence of a recombinantly expressed protein selectively recognized by targeted transgenic T cells. Transgenic and wildtype (WT) T-cells were isolated and co-cultured at 1:1, 5:1, and 10:1 T-cell:target-cell ratios using LTFs from both tumor types and assessed for T-cell activation and tumor cell viability using flow cytometry and cytokine/chemokine secretion profiling.

TILs: Tumors were collected and processed for TIL expansion, which was conducted in the presence of IL-2 for 10 days. Following expansion, CD8 T-cells were isolated and co-cultured for 48h at a 1:1 ratio with LTFs from the same tumor. Tumor killing and T-cell activation were measured by flow cytometry.

All animal studies were carried out in accordance with animal welfare guidelines and approved by the IACUC at Excelsior Labs.

Results Transgenic T-cells: Flow cytometry revealed an increase in activation markers (CD25, CD69, ICOS, PD1, and CD137) on the surface of transgenic CD8 T-cells incubated with target-positive tumors, but not target-negative tumors, while WT T-cells displayed no increases when incubated with either tumor type. Secretome profiling demonstrated similar changes, with an increase in cytokine and chemokine secretion (IFNg, CXCL1, CCL4, and CXCL5) as the ratio of transgenic CD8 T-cells to target-cells increased, using target-positive, but not target-negative, LTFs. Decreases in tumor cell viability were also detected at higher T-cell:Target-cell ratios and were again only observed with target-positive LTFs.

TILs: CD8 T-cells isolated from TILs, but not WT T-cells, that were co-cultured with LTFs demonstrated increased activation and resulted in a decrease in tumor cell number following 48h of culture.

Conclusions We have verified target-specific transgenic T-cell activation, as well as reduced tumor viability in expanded TILs following co-culture with LTFs. These data lay the groundwork for future studies using the Elephas LTF platform to predict individual clinical responses to various T-cell therapies.

Ethics Approval All animal studies were carried out in accordance with animal welfare guidelines and approved by the IACUC at Excelsior Labs.