Background Adoptive cell therapy with genetically modified T cells has shown promising efficacy in solid tumors but has been limited by immunosuppressive mechanisms that interfere with sustained activity including FAS ligand-induced apoptosis of tumor-infiltrating, FAS receptor-positive lymphocytes. As previously reported, T Cell Receptor (TCR)-engineered T cells expressing a FAS-41BB switch receptor, consisting of FAS extracellular and 41BB intracellular domains, demonstrated improved anti-tumor efficacy. KRAS is a frequently mutated oncogene in cancers and recent clinical evidence suggests that it is immunogenic and targetable via TCR-engineered T cells. Targeting a mutated oncogenic driver such as KRAS G12V offers many advantages, including tumor dependence driving homogenous expression and decreasing risk of therapeutic escape. We are now reporting an optimized construct that achieves high functional co-expression of the KRAS TCR, CD8ab, and FAS-41BB switch receptor in a single viral vector.

Methods Human T cells isolated from healthy volunteers were lentivirally transduced with constructs encoding the KRAS TCR, CD8ab chains, and FAS-41BB. Preclinical studies included peptide titrations with the index peptide and ones in which one residue was individually substituted to all possible amino acids (XScan), co-cultures with tumors or B-LCL, and in vivo subcutaneous xenografts.

Results Co-expression of CD8ab with the KRAS G12V-specific TCR allowed for efficient stimulation of CD4+ T cells, and further engineering with the FAS-41BB switch receptor increased sensitivity to low peptide concentrations. In tumor co-culture assays, the inclusion of FAS-41BB allowed for tumor control, even after re-challenge with fresh addition of tumor cells. In a repetitive T cell transfer/tumor exposure assay, continued proliferation and tumor control required both expression of FAS-41BB and inclusion of engineered CD4 and CD8 T cells, suggesting that CD8ab exogenous expression in CD4 T cells allowed for a coordinated T cell response able to resist exhaustion. Intravenous administration of engineered T cells prevented tumor outgrowth in vivo. Using the XScan assay, no off-target liabilities were identified upon co-incubation of A11-KRAS G12V/CD8ab/FAS-41BB switch receptor engineered T cells with all possible peptides in the human proteome matching the recognition motif, demonstrating the specificity of our TCR. No alloreactivity to the most prevalent HLA alleles was detected in B-LCL co-cultures.

Conclusions Preclinical development of the KRAS G12V-specific TCR with co-expression of CD8ab and the durability FAS-41BB switch receptor supports the clinical development of this first-in-class product for solid tumor patients with high unmet medical needs.

REFERENCES