Background: Adoptive cell therapy based on chimeric antigen receptor-engineered T (CAR-T) cells has proven to be life-saving for many cancer patients. However, its therapeutic efficacy has so far been restricted to only a few malignancies, with solid tumors proving to be especially recalcitrant to efficient therapy. Poor intra-tumor infiltration by T cells and T cell dysfunction due to an immunosuppressive microenvironment are key barriers against CAR-T cell success against solid tumors. Furthermore, low level expression of CAR-directed tumor-associated antigens (TAA) in normal tissues can result in “on-target off-tumor” cytotoxicity, raising potential safety concerns.

Methods: Using our TALEN®-based gene editing platform, we present here innovative T cell engineering strategies that can combat some of the challenges posed by CAR-T cell development for solid tumors. These allogeneic ‘Smart CAR-T’ cells are designed to integrate locus-specific synthetic genes that can either respond to or take advantage of the unique cues localized to the solid tumor microenvironment (TME).

Results: Inducible expression of a tumor-antigen directed CAR by a constitutive CAR specific to TME cues greatly enhanced anti-tumor activity, while limiting ‘on target, off-tumor’ cytotoxicity. Additionally, CAR induced gene expression could boost anti-tumor CAR-T only within the TME. Thus, our gene editing strategies could increase CAR-T cell persistence and anti-tumor activity while staying restricted to the tumor milieu.

Conclusions: Our proof-of-concept study demonstrates the feasibility of developing CAR-T cell engineering strategies that can improve solid tumor targeting while mitigating potential safety risks, paving the way for clinical development.

Ethics Approval: All procedures involving animals were performed in accordance with regulations and established guidelines and were reviewed and approved by the Cellectis Institutional Animal Care and Use Committee (IACUC), as well as by the Animal Ethical Committee at Mispro-Biotech (New York, NY)