Supplemental Information

Intratumoral injections of chimeric antigen receptor (CAR) T cells directed against c-Met are safe and induce tumor necrosis and inflammatory responses in metastatic breast cancer

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Supplemental Materials

Supplementary Figure Legends

Fig. S1. Clinical trial schema. Design of a phase 0 clinical trial (NCT01837602) to evaluate the safety and feasibility of intratumoral injection with mRNA c-Met-CAR T cells in patients with metastatic breast cancer.

Fig S2. Representative immunohistochemistry (IHC). Two breast cancer tumor samples displaying intratumoral heterogeneity in intensity and proportion of tumor cells demonstrating membrane staining of c-Met on IHC. A) 1+ 30% c-Met positive breast cancer. Note: about 30% of tumor cells had membranous c-Met staining while remaining tumor cells had background c-Met staining; and B) 3+ 90% c-Met positive breast cancer. Note: the majority of tumor cells were strongly (3+) c-Met+ while stromal cells were c-Met negative.

Fig. S3. Histology and IHC of tumor tissue pre and post intratumoral injection of RNA CAR T c-Met from one patient. Histologic (H&E) and IHC evaluation of c-Met expression and T cell (CD3, CD4, CD8) and macrophage (CD68) infiltration in tumor tissues of pre-treatment, away from intratumoral injection site and at intratumoral injecton site.

Fig. S4. Histology and IHC of tumor tissue pre and post intratumoral injection with lidocaine (1 mL) and RNA CAR T c-Met (1 mL) from another patient. Histologic (H&E) and IHC evaluation of c-Met expression and T cell (CD3, CD4, CD8) and macrophage (CD68) infiltration in tumor tissues of pre-treatment, intratumoral injection of lidocaine site and intratumoral injection of mRNA c-Met-CAR T cells.

Fig. S5. Characterization of CD68+ tumor infiltrated immune cells using multiplex IHC analyses as described (1) **in pre and post IT injection of RNA CAR T c-Met tumor tissue sections from two patients.** The percentages of immune cells staining positive for the following markers (CD66b, CD68, CD14, CD163, and HLA-DR) in pre-treatment (black bars) and IT injection site (grey bars) were depicted.

Supplemental Reference

1. Tumeh PC, Harview CL, Yearley JH, Shintaku IP, Taylor EJM, Robert L, et al. PD-1 blockade induces responses by inhibiting adaptive immune resistance. Nature. 2014 Nov 27;515(7528):568–71.