**Supplementary Materials and Methods**

**Antibodies*.*** Purified anti-mouse CD3ε (145-2C11), anti-mouse CD28 (37.51), and anti-mouse CD16/32 (Clone 93) antibodies were purchased from eBioscience. Anti-CD45 (Clone 30-F11; PE-Cy7), anti-CD90 (Clone 53-2.1; FITC, PE) anti-F4/80 (clone BM8; APC) and anti-CD206 (clone C068C2; FITC) antibodies were purchased from Biolegend. APC-conjugated anti-CD8α (Clone 53-6.7) was purchased from BD Bioscience.Polyclonal sheep anti-human antibody that cross-reacts with mouse FAP was purchased from R&D and the specificity was verified based on reactivity with tumors derived from wild-type but not FAP-null mice.

**Synthesis of anti-muFAP CAR constructs.** The anti-mouse FAP-CAR construct containing a single-chain Fv domain of the anti-mouse FAP antibody (73.3 scFv) and the CD3 and 4–1BB intracellular signaling domains was engineered as described (ref. 44). The efficacy of this construct was demonstrated in multiple preclinical flank tumor mouse models (ref. 44).

**Isolation, Transduction and Expansion of Primary Mouse T lymphocytes.** Murine splenic T cells were isolated using the “Pan T cell Negative Selection” kit (Miltenyi Biotec), and incubated in 24-well plates (4×106 cells/well in 2 mL supplemented RPMI-1640 with 100 U/mL IL-2) pre-coated with -CD3 (1 g/mL) and -CD28 (2 g/mL). After 48 hours, cells (1x106 cells/well) were mixed with retrovirus (1 mL crude viral supernatant) in a 24-well plate coated with Retronectin (50 g/mL; Clontech) and centrifuged, without braking, at room temperature for 45 minutes at 1200g. After overnight incubation, cells were expanded with 50 U/mL of IL-2 for additional 48 hours

**Lentivirus preparation.** The anti-mouse FAP-CAR was subcloned into a third generation self-inactivating lentiviral expression vector (pTRPE), in which transgene expression is driven by the EF-1 promoter. High-titer replication-defective lentiviral vectors were produced and concentrated as described (Suppl. ref 1).

**Isolation, transduction, and expansion of primary human T lymphocytes.** Human T cells expressing CAR were prepared as described (Suppl, ref. 2). All specimens were collected under a University Institutional Review Board-approved protocol, and written informed consent was obtained from each donor. T cells were cultured in RPMI 1640 supplemented with 10% FCS, 100 U/ml penicillin, 100g/ml streptomycin sulfate, 10 mM HEPES, and stimulated with magnetic beads coated with anti-CD3/anti-CD28 (Invitrogen) at a 3:1 bead to cell ratio. Approximately 24 h after activation, T cells were transduced with lentiviral vectors at an MOI of 5. Cells were counted and fed with fresh culture medium containing 30U/mL of IL2 every 2 days and once T cells appeared to rest down, as determined by both decreased growth kinetics and cell size, they were either used for functional assays or cryopreserved.

**Flow cytometric analyses.** Single cell suspensions were prepared from tumors cut into fragments and digested with collagenase type I (100U/ml, Worthington), II (100U/ml, Worthington) and IV (100U/ml, Worthington), DNase I (100 U/ml, Worthington). Cell acquisition was performed on LSR-II using FACSDiva software (BD Bioscience, USA). Data were analyzed using FlowJo (Tree Star).

**Histopathological, histochemical and immunohistochemical analysis**. Tumors were harvested, fixed with Prefer (Anatech), dehydrated and paraffin embedded. Tissue sections were deparaffinized, rehydrated and stained with Masson’s tricrhome, alcian blue or H&E. For immunohistochemical staining, primary antibodies **(Supplementary Table 1)** were applied overnight at 4°C and incubated with biotinylated secondary antibody for 1 hr at RT. Bound antibodies were detected with horseradish peroxidase (HRP), using either streptavidin-HRP (Jackson ImmunoResearch) or Vectastain Elite ABC (Vector Laboratories) and counterstained with hematoxylin. Digital images were captured using a Nikon E600 microscope at high power field (HPF) (20X), at least 5 images per sample. Quantification was performed using Fiji software. Proliferation index or apoptotic index was calculated by the ratio of ki-67+ cells or Cleaved-caspase-3+ cells to hematoxylin+ cells. Histochemical-reactive or immuno-reactive signals were calculated and normalized to total area of HPF images (n= 3 to 6 per group).

**Supplementary reference**

1. Moon EK, Carpenito C, Sun J, Wang LC, Kapoor V, Predina J, et al. Expression of a functional CCR2 receptor enhances tumor localization and tumor eradication by retargeted human T cells expressing a mesothelin-specific chimeric antibody receptor. Clin Cancer Res 2011;17:4719-30.
2. Parry RV, Rumbley CA, Vandenberghe LH, June CH, Riley JL. CD28 and inducible costimulatory protein Src homology 2 binding domains show distinct regulation of phosphatidylinositol 3-kinase, Bcl-xL, and IL-2 expression in primary human CD4 T lymphocytes. J Immunol 2003;171:166-74.