Supplemental Figure 1. A capacity of tumor-infiltrating anti-FITC CAR T cells to produce cytokines/chemokines in response to FITC stimulation.

SW480 human colon cancer cells were implanted s.c. into NSG mice followed by injection with FITC-Ctx or unlabeled Ctx one day later. Anti-FITC CAR T cells were injected i.v. one day after Ab injection. Thirty days after T cell transfer, mice were euthanized and T cells were extracted from the tumors, followed by in vitro re-stimulation with irradiated SW480 cells in the presence of FITC-Ctx or unlabeled Ctx. Three days later, production of indicated cytokines and chemokines was examined by Milliplex assay.
Supplemental Figure 2. Functional role of human CD3ζ/CD28/4-1BB signaling motifs in mouse CAR T cell activation. Mouse T cells were transduced with retrovirus vector encoding anti-FITC CAR (CAR) or anti-FITC CAR lacking intracellular human CD3ζ/CD28/4-1BB signaling motifs (ΔCAR). These T cells were cultured with plate-coated FITC-Rtx (+) or non-labeled Rtx (-). After 72 hrs, the culture supernatants were harvested and production of indicated cytokines and chemokines were measured by multiplex assay. Data are shown as mean ± SD.
Supplemental Figure 3. Development of anti-FITC Ab responses in the mice injected with FITC-conjugated Ab.
C3H/HeN mice inoculated with human CD20-positive 38C13 lymphoma were treated i.v. with anti-FITC CAR T cells plus i.p. injections of either FITC-labeled Rtx (●) or non-labeled Rtx (□), as indicated in Figure 6A. After 20 days, mouse serum was harvested, diluted as indicated, and tested for a binding to plate-coated FITC-dextran by ELISA. Data (O.D.: optical density) is shown as mean ± SEM from at least 3 mice per group.