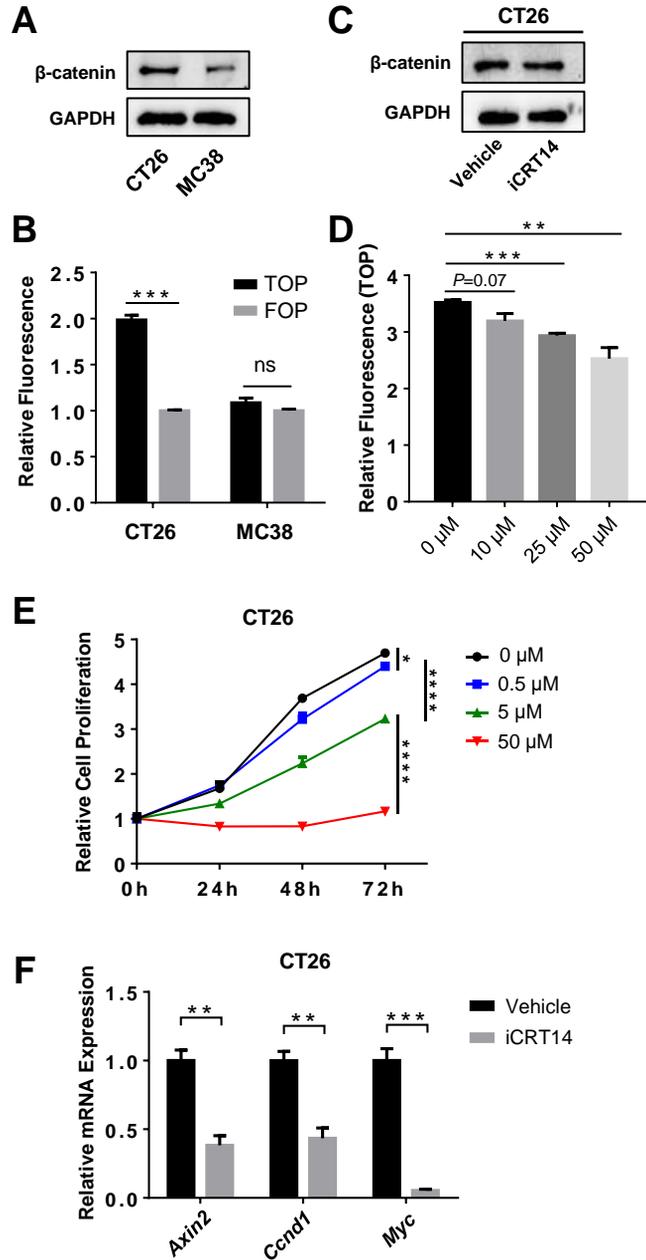
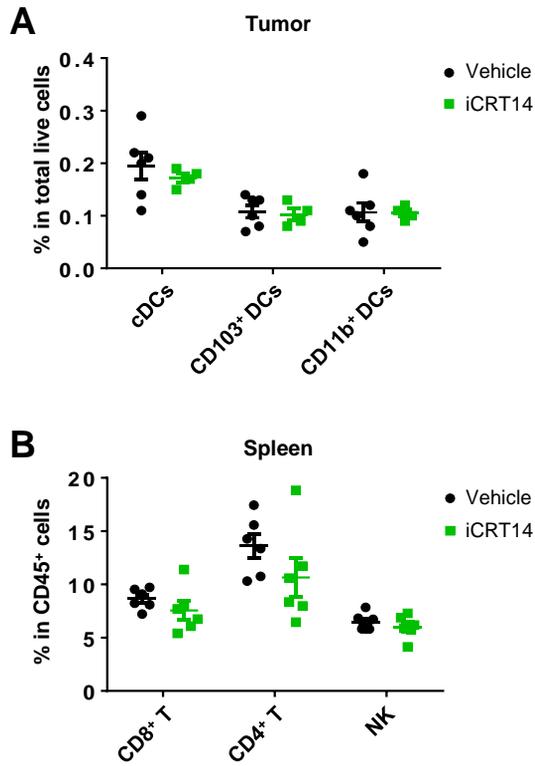


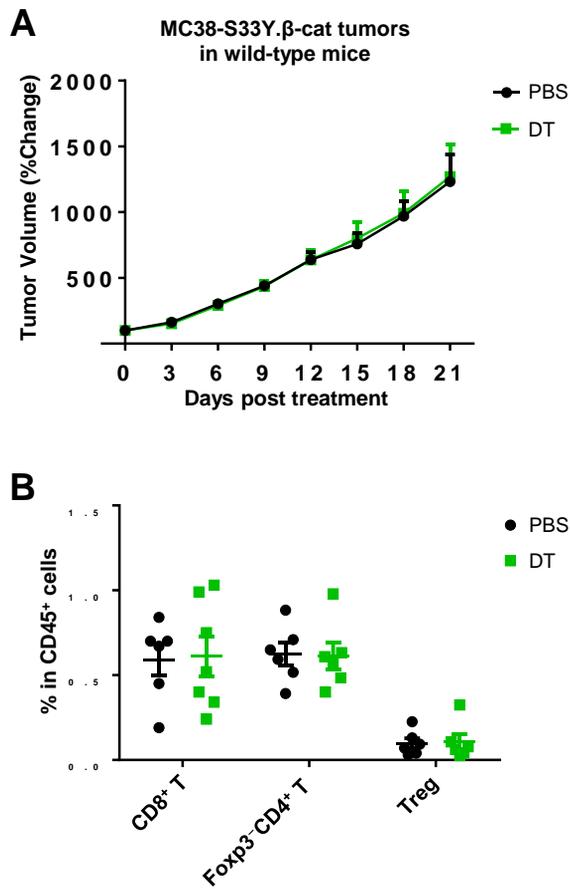
Supplemental Figure S1. iCRT14 inhibits the activity of mutant β -catenin. **(A)** 293T cells were transfected with β -catenin/TCF reporter construct (TOPflash) and its mutant control (FOPflash). Cells were treated with 10 nM LY2090314 and iCRT14 at indicated concentrations. $n = 4$. **(B)** 293T cells were transfected with TOPflash together with empty vector, S33Y. β -catenin or Δ 45. β -catenin constructs, then treated with 25 μ M iCRT14 or 25 μ M XAV939 for 24h. $n = 4$. **(C)** HCT116 cells were treated with 25 μ M iCRT14 or 25 μ M XAV939 for indicated time. Viable cells were quantified by CCK-8 assay. $n = 4$. *** $P < 0.001$, **** $P < 0.0001$.



Supplemental Figure S2. iCRT14 inhibits colorectal tumor cell growth and target gene expression. (A) Western blot for β -catenin and GAPDH in CT26 and MC38 cells. (B) Activities of β -catenin/TCF reporter (TOP) or its mutant control (FOP) in CT26 and MC38 cells. $n = 3$. (C) Western blot for β -catenin in CT26 cells treated with 25 μ M iCRT14 for 24 hours. (D) Activity of β -catenin/TCF reporter (TOP) in CT26 cells treated with iCRT14 at indicated concentrations. $n = 3$. (E) CT26 cells were treated with 0 μ M, 0.5 μ M, 5 μ M or 50 μ M iCRT14 for indicated time. Viable cells were quantified by CCK-8 assay. $n = 4$. (F) Quantification of mRNA expression by qPCR. $n = 3$. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, **** $P < 0.0001$.



Supplemental Figure S3. iCRT14 treatment does not affect tumor-infiltrating dendritic cells or splenic immune cells. Mice bearing CT26 tumors were treated with iCRT14 or vehicle for 12 days. FACS was performed to analyze tumor-infiltrating cDCs (CD45⁺CD11c⁺MHC-II⁺CD64⁻Gr-1⁻) (A), and splenic T and NK cells (B).



Supplemental Figure S4. DT treatment does not affect tumor growth or T cell infiltration in wild-type mice. Wild-type C57BL/6 mice bearing MC38-S33Y. β -cat tumors were treated with DT or PBS every four days. Tumor growth was monitored for 21 days post the first injection ($n = 8/\text{group}$) (A). FACS was performed one day after the third injection of DT to analyze tumor-infiltrating CD8⁺ T, conventional Foxp3⁻CD4⁺ T and Treg cells (B).