



Supplemental Figure S5: In vivo TR-APC induction drives oligoclonal T cell expansion. **A**, Schematic of TCRV β usage analysis. Peripheral blood draws on Day 0, prior to tumor inoculation were used to enumerate baseline TCRV β frequencies in each animal by flow cytometry. Animals were engrafted with RAW-112 cells and TR-APCs were induced by doxycycline chow. 14 days after tumor inoculation, peripheral blood draws were again used to enumerate TCRV β frequencies in each animal. **B**, Representative gating scheme for enumerating TCRV β frequencies. Cells from each peripheral blood draw were divided evenly into 15 groups and stained with an antibody

cocktail containing 1 of 15 individual α -TCRVb antibodies. TCRVb⁺ cells were enumerated as a frequency of single, live, CD3⁺CD4⁺/CD8⁺ cells. **C**, Raw frequency of each TCRVb population among CD8⁺ T cells from each animal at baseline (left bar of each animal) and Day 14 post tumor injection (right bar of each animal). Mice #120-124 received doxycycline chow and mice #125-129 received normal chow. **D**, Change in frequency of each TCRVb population of CD8⁺ T cells for each mouse in **A-B**. Change in frequency was calculated as (final frequency - initial frequency) and is plotted here as a superimposed bar graph. **E**, Raw frequency of each TCRVb population among CD4⁺ T cells from each animal at baseline (left bar of each animal) and Day 14 post tumor injection (right bar of each animal). Mice #120-124 received doxycycline chow and mice #125-129 received normal chow. **F**, Change in frequency of each TCRVb population of CD4⁺ T cells for each mouse in **A-B**. Change in frequency was calculated as (final frequency - initial frequency) and is plotted here as a superimposed bar graph. n=5 mice/group. *P* values for TCRVb 14 frequency were calculated with two way ANOVA with Sidak's multiple comparison test. **** $P_{\text{adj}} \leq 0.0001$, ** $P_{\text{adj}} \leq 0.01$.