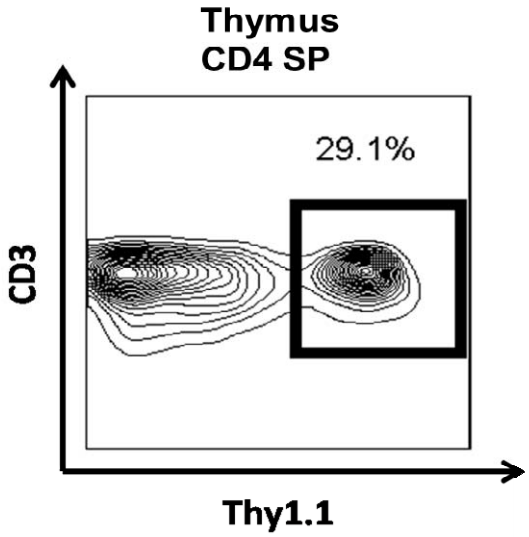

Figure S1. CA30 thymocytes have competitive disadvantage relative to wild type thymocytes. Thymus of control mixed BMC generated with equal numbers of Thy1.1 CA30 and Thy1.2 B6AF1 donor bone marrow cells. $CD4^+$ SP thymocytes were segregated on the basis of the Thy1.1 marker. Representative of data from 3 mice.

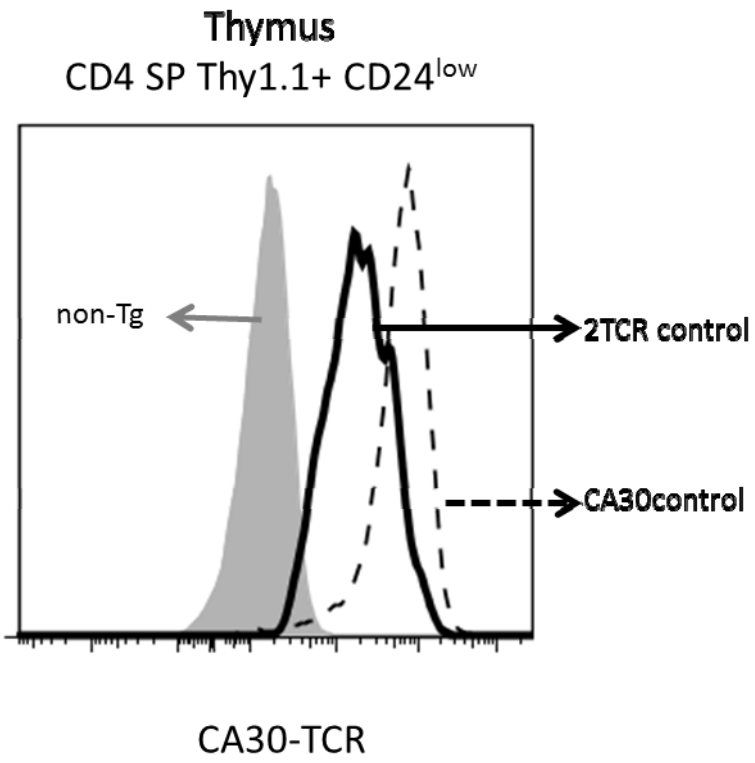

Figure S2. Mature 2TCR thymocytes express low levels of the CA30 TCR. Stain of CD4-SP Thy1.1^{+}CD24^{low} thymocytes from 2TCR (thick line) and CA30 (dashed line) mice with with Vκ36-71 FR1-I-A^{k} tetramer. NonTg mature thymocytes were used as a negative control.

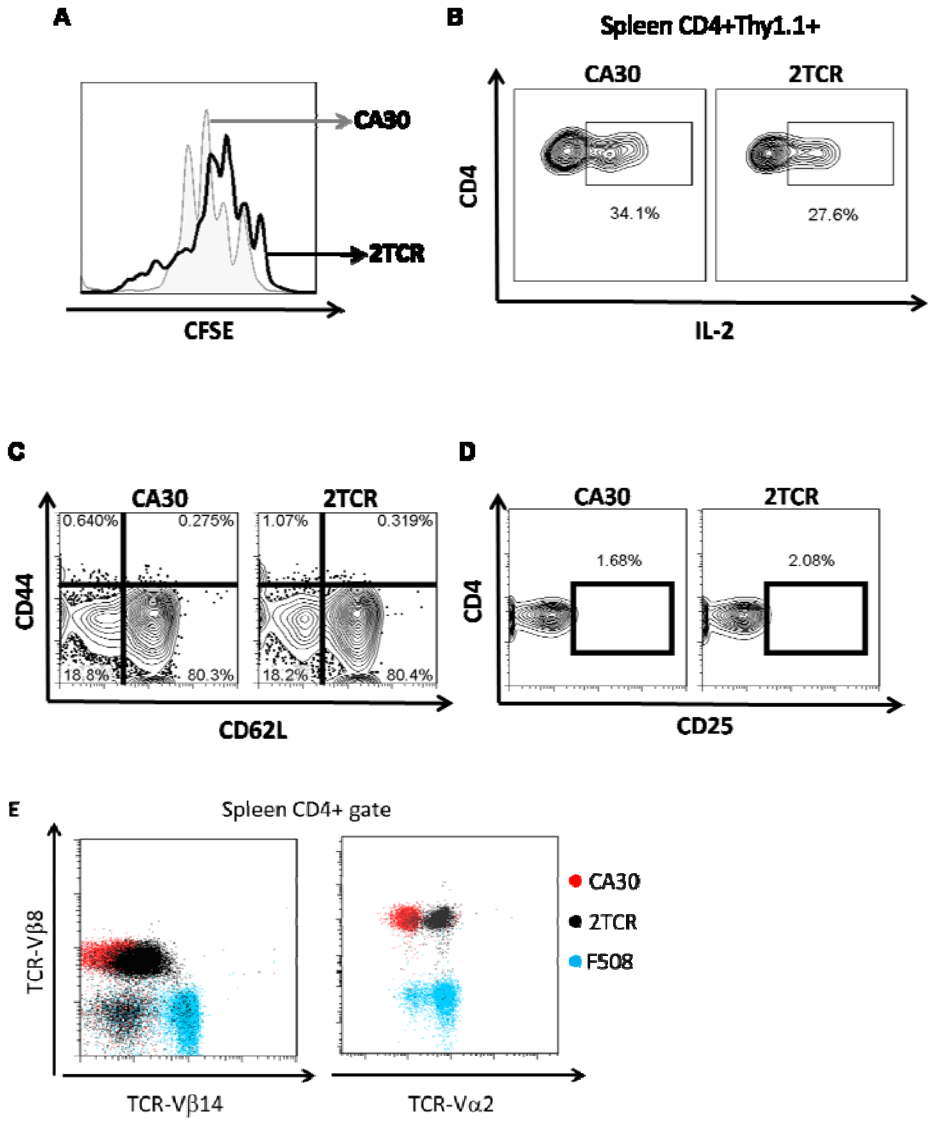

Figure S3. Diminished proliferative response to Vk36-71 FR1 peptide by 2TCR T cells. *A*, Purified CA30 and 2TCR T cells were CFSE labeled and incubated with 100 nM of Vκ36-71 FR1 peptide for 5 days. *B,* CA30 or 2TCR T LN cells (10^6) were transferred into B6AF1 mice, which were immunized with the Vκ36-71 FR1 peptide (10 μg) in (7 μg) LPS. At day 9, *ex vivo* splenocytes were cultured (10^6/ml) with Vκ36-71 FR1 peptide (1 μM) for 6h and stained for intracellular IL-2. *C-D,* Expression of CD44, CD62L and CD25 by $CD4^+$ splenocytes from CA30 and 2TCR mice. *E,* Expression of TCR-Vβ8, TCR-Vβ14 and TCR-Vα2 by $CD4^+$ splenocytes from CA30 and 2TCR Tg mice.

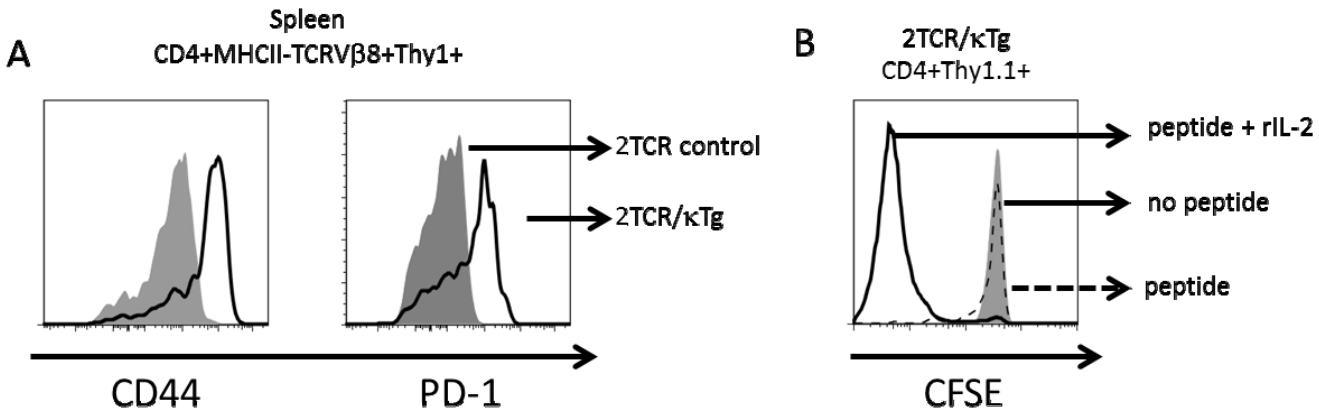

Figure S4. 2TCR T cells in 2TCR/κTg chimeras have an antigen experienced phenotype and are anergic *in vitro*. *A,* Expression of activation markers by 2TCR $CD4^+$ T cells in 2TCR/κTg (thick line) versus control 2TCR/B6AF1 (solid histogram) chimeras. *B, In vitro* proliferation of 2TCR T cells derived from 2TCR/κTg chimeras only when rIL-2 is provided. Cells were cultured with Vκ36-71 FR1 peptide (1 μM) alone or with rIL-2 (50 U/ml).