Supplemental Table 1. Primers used in this study.

Genotyping		
RARα-Tg	Forward	GATGCTGATGAAGATCACAG
	Reverse	GGACAATGAGTTTTCTGCTG
Internal control (RARα-Tg)	Forward	TCAGCCCCTCACCCTCCAAT
	Reverse	CTCACCTTACAGCCCTCACA
Cre	Forward	CGGTCGATGCAACGAGTGATGAGG
	Reverse	CCAGAGACGGAAATCCATCGCTCG
Internal control (Cre)	Forward	CTAGGCCACAGAATTGAAAGATCT
	Reverse	GTAGGTGGAAATTCTAGCATCATCC
Rara fl/fl	Forward	TCAGCCCCTCACCCTCCAAT
	Reverse	CTCACCTTACAGCCCTCACA
qRT-PCR		
Rara	Forward	CCTGCCCCGCATCTACAAG
	Reverse	GGTTCCGGGTCACCTTGTT
Slc2a1 (Glut1)	Forward	GTGACGATCTGAGCTACGGG
	Reverse	ACTCCTCAATAACCTTCTGGG
Glud1	Forward	GCATCTTGGAGGCTGACTGT
	Reverse	GCACCCGATATCCTGTCCTG
Slc7a5 (Lat1)	Forward	ATCGTGGGCACCATCATCG
	Reverse	CAGCTGTGAGGAGCAGCA
Icos	Forward	TCATCTGTCAAACAACAGCGTC
	Reverse	AGGTCACACCTGCAAGTCTAGA
Actb	Forward	AGAAGAGCTACGAGCTGCCTGAC
	Reverse	TACTCCTGCTTGCTGATCCACAT

Supplemental Figures

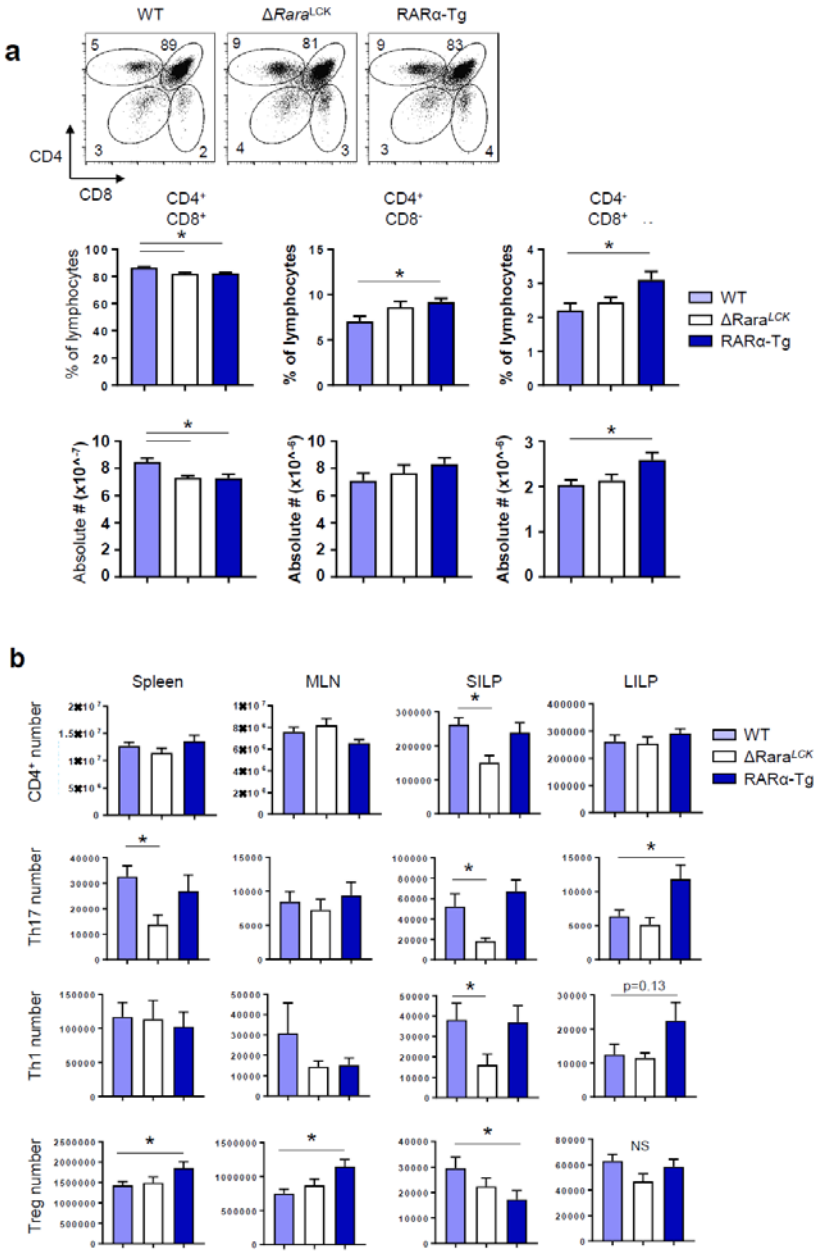

Fig. S1. T lymphopoiesis and peripheral CD4⁺ T subsets in WT, ΔRara^{LCK}, and RARα-Tg mice. (a) Thymic T cell populations in 6-8 week-old mice. (b) Total T helper, Treg, Th17, and Th1 cell numbers in spleen, MLN, small intestinal lamina propria (SILP), and large intestinal lamina propria (LILP) of WT, ΔRara^{LCK}, and RARα-Tg mice. Representative and combined data (n=6-7) from at least 3 independent experiments are shown. All error bars indicate SEM. *Significant differences (*P* values <0.05) between indicated groups as analyzed by one-way ANOVA with Bonferroni.

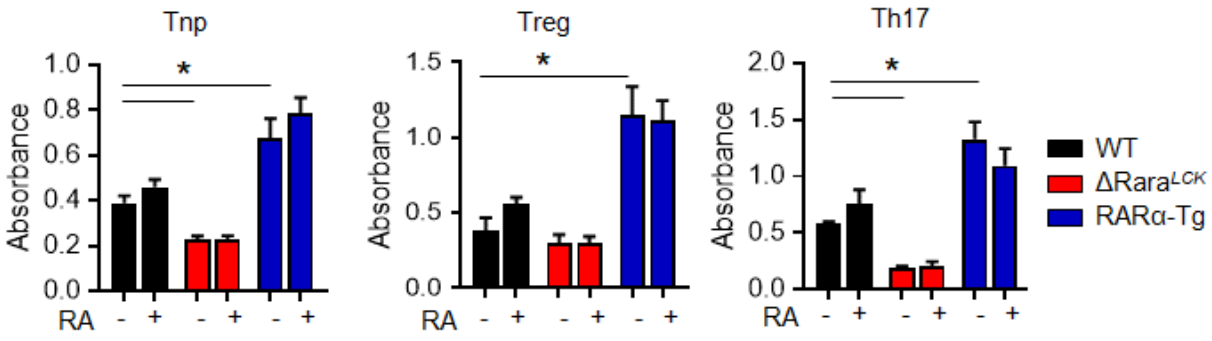

Fig. S2. Proliferation of WT, ΔRara^{LCK}, and RARα-Tg cells. XTT staining levels of cultured CD4$^+$ T cells indicating cell proliferation at 72h. Combined data from 3 independent experiments are shown. All error bars indicate SEM. *Significant differences (*P* values <0.05) between indicated groups as analyzed by two-way ANOVA with Bonferroni.

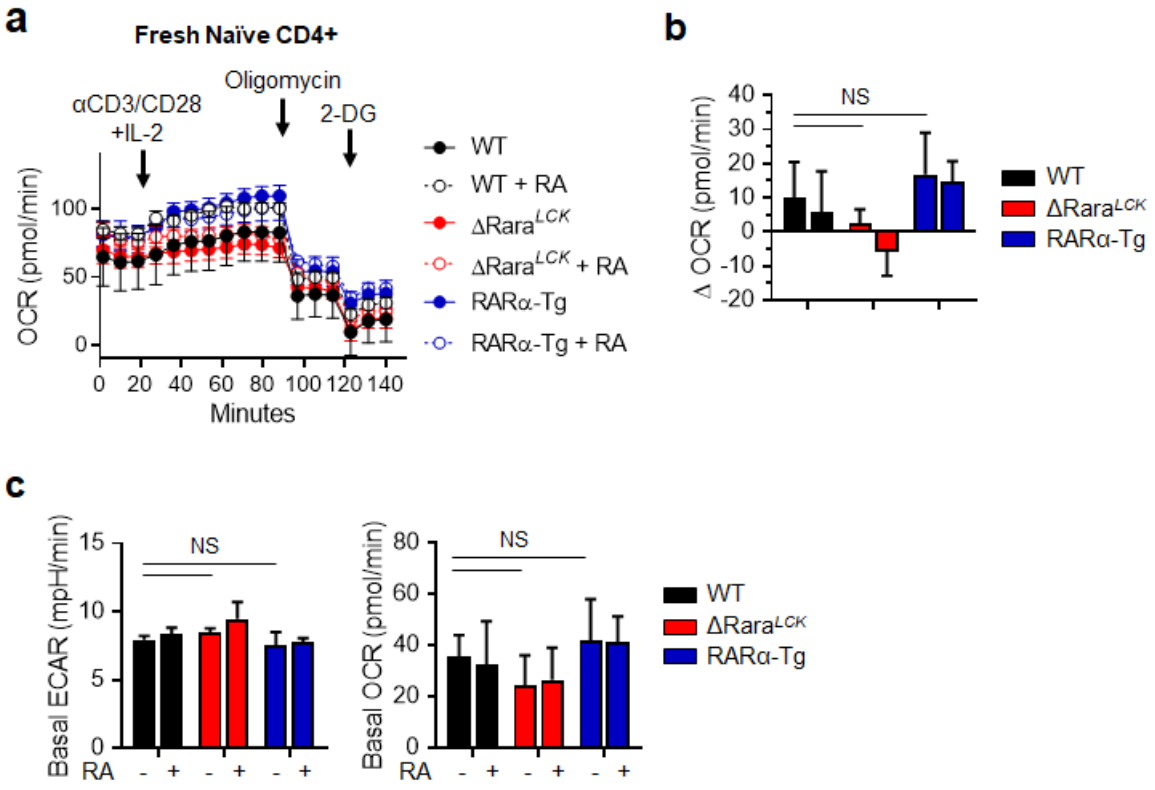

Fig. S3. Impact of the RARα expression level on mitochondrial metabolism and basal ECAR and OCR. (a) Seahorse assay OCR measurements of naïve CD4$^+$ T cells upon anti-CD3/CD28 and IL-2 stimulation. (b) Changes in OCR (ΔOCR) during the first ~70 min period following activation. (c) Basal ECAR and OCR rates of naïve T cells prior to activation. Representative and combined data (n=3 for a-c) are shown. All error bars indicate SEM. *Significant differences (*P* values <0.05) from control or between two groups analyzed by repeated-measures two-way ANOVA with Bonferroni.

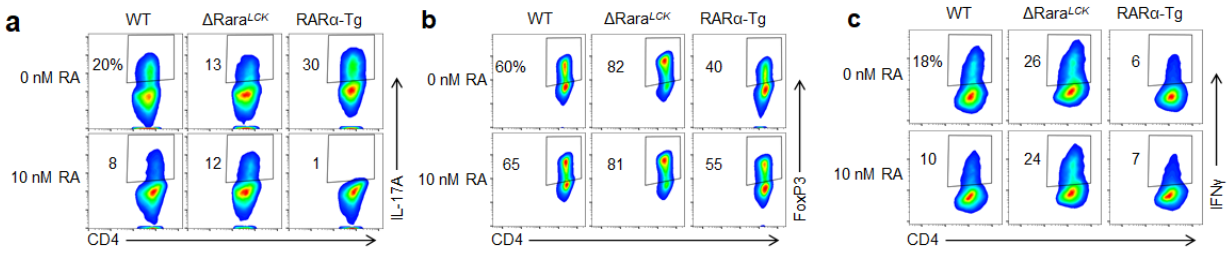

Fig. S4. Impact of RARα dose on Th cell polarization *in vitro*. Naïve CD4$^+$ T cells, isolated from indicated mouse lines were cultured in a Th17 (a), Treg (b), or Th1 (c) polarization condition for 4 days. Representative flow dot plots are shown (n=3-5).

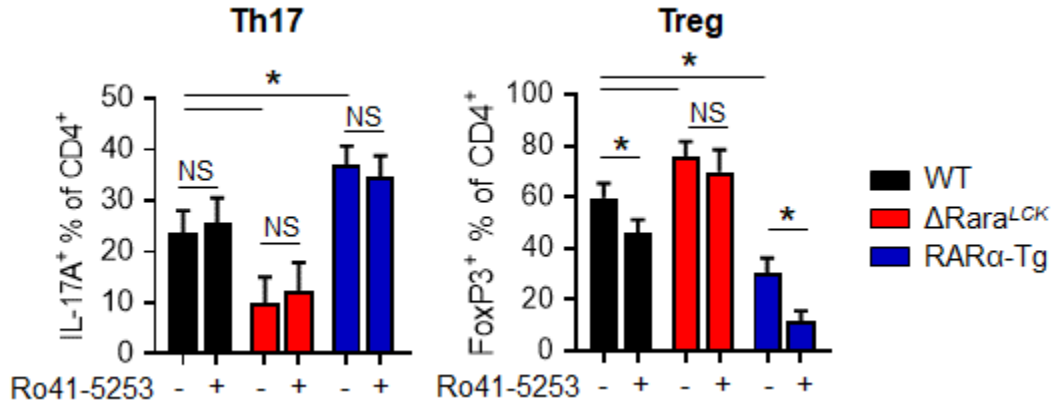

Fig. S5. Impact of a RARα antagonist (Ro41-5253) on Th17/Treg polarization in the low RA condition with charcoal-treated FBS. Naïve CD4$^+$ T cells, isolated from indicated mice, were cultured for 4 days in a Th17 or Treg polarizing condition in the presence or absence of Ro41-5253 (100 nM). All error bars indicate SEM. *Significant differences (*P* values <0.05) from control or between two groups analyzed by repeated-measures two-way ANOVA with Bonferroni.

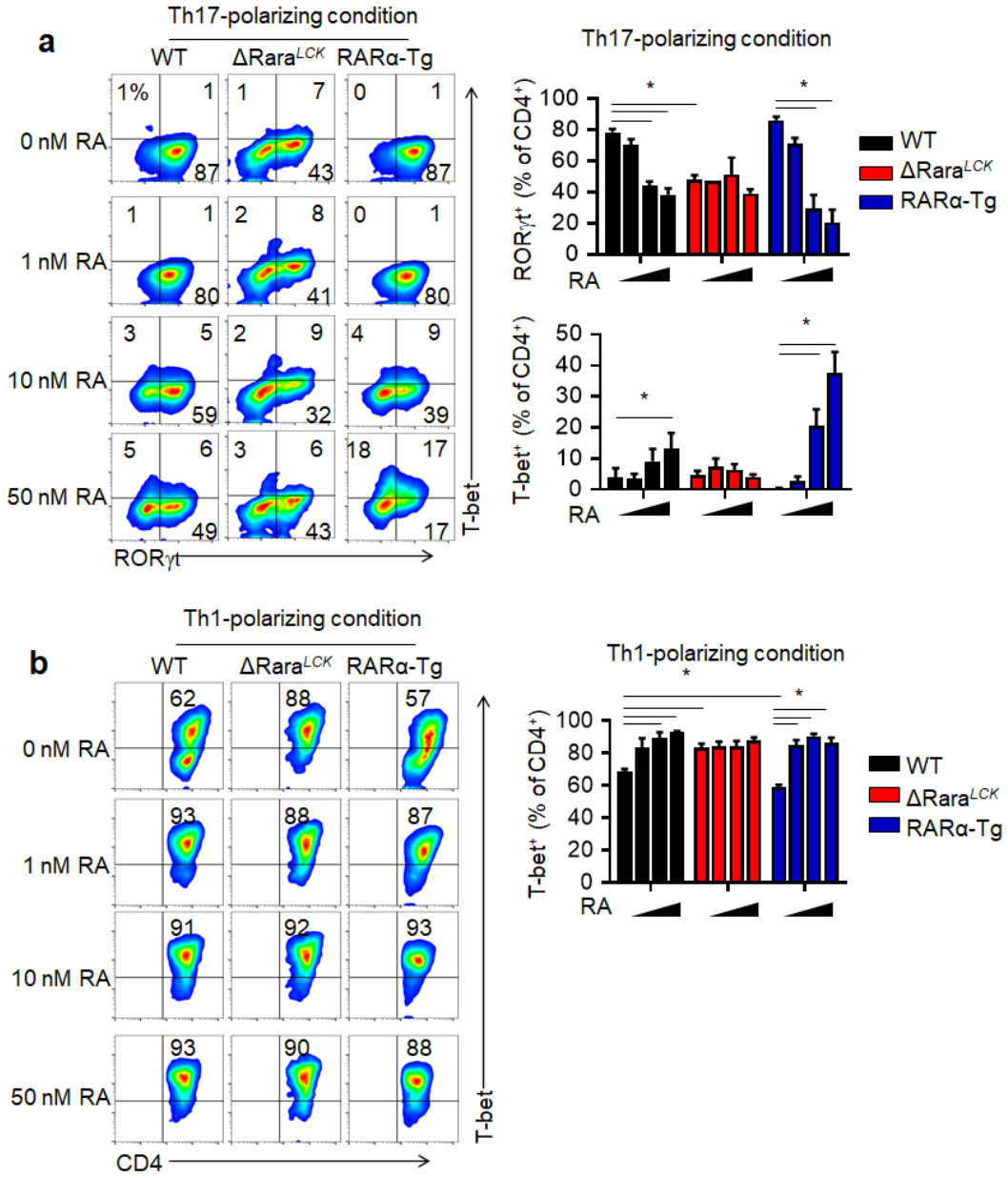

Fig. S6. Impact of RARα dose on Th cell polarization *in vitro*. Naïve CD4$^+$ T cells, isolated from indicated mouse lines were cultured in a Th17 (a) or Th1 polarization condition for 4 days. (a) RORγt and T-bet expression at day 4 in culture. (b) T-bet expression at day 4. Representative flow dot plots are shown (n=3). All error bars indicate SEM. *Significant differences (*P* values <0.05) from control or between two groups analyzed by repeated-measures two-way ANOVA with Bonferroni.

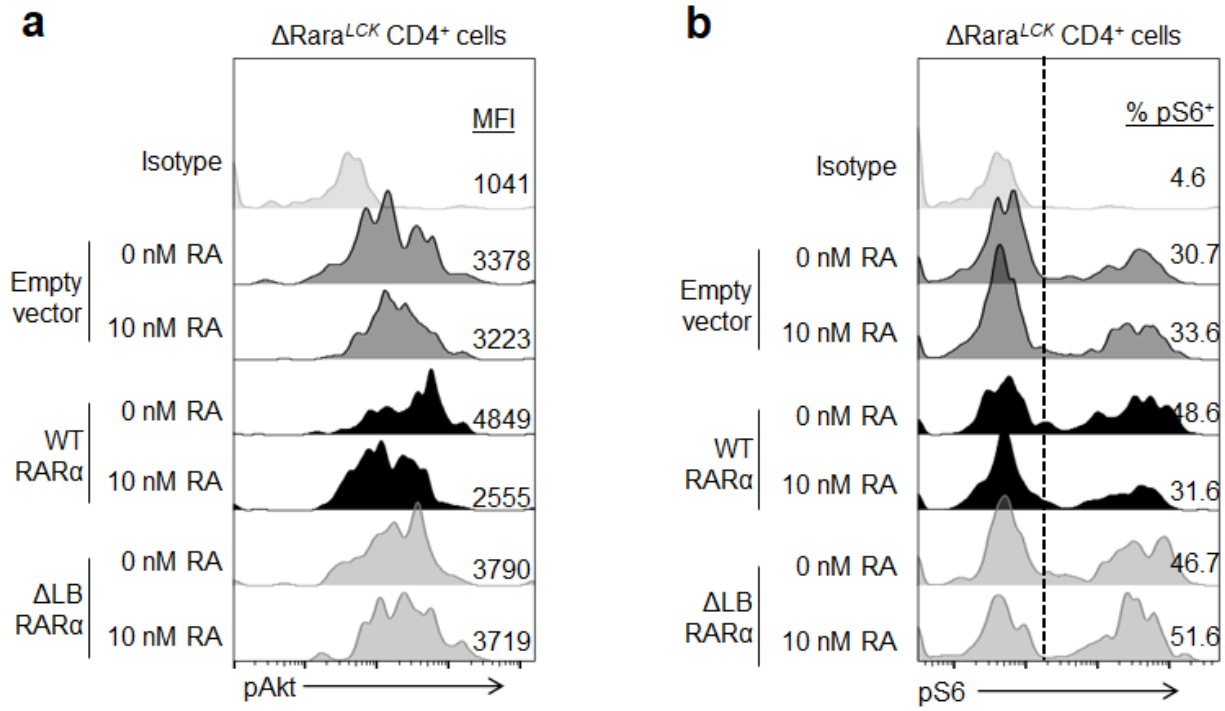

Fig. S7. Impact of RARα restoration on Akt and mTOR activity. Naïve ΔRara^{LCK} CD4 T cells were transduced with retroviral vectors. (a, b) Cells were first cultured overnight with anti-CD3/CD28 activation, transduced, and rested for 24 h prior to reactivation with anti-CD3/CD28 and hIL-2 and flow cytometry analysis. Representative flow cytometry histograms are shown.

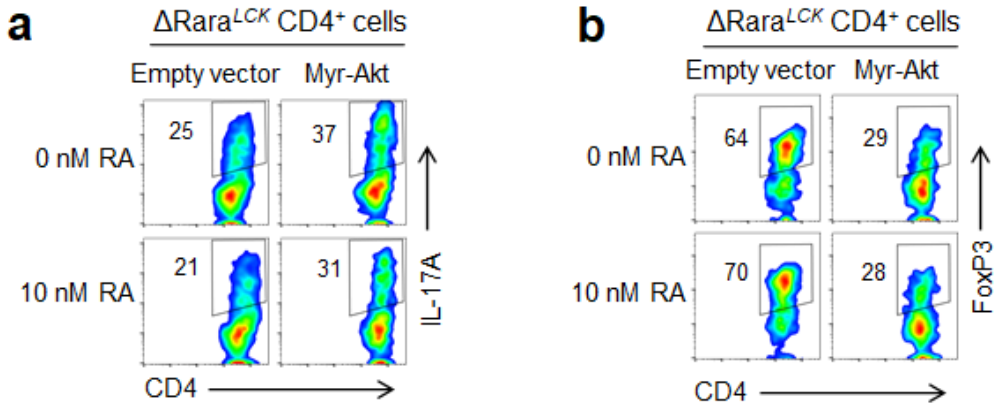

Fig. S8. Impact of constitutively active Akt activity on Th17/Treg polarization. Naïve ΔRara^{LCK} CD4 T cells were transduced with retroviral vectors. Cells were first cultured overnight with anti-CD3/CD28 activation, transduced, and cultured for 3 additional days in Th17 (a) or Treg (b) inducing condition. Representative flow cytometry dot plots are shown.

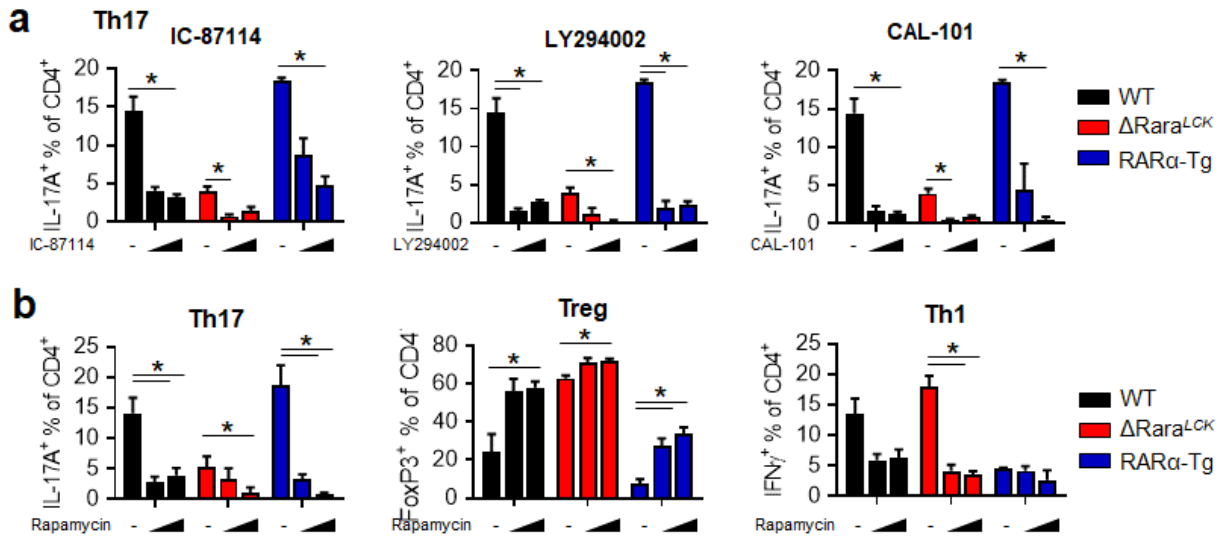

Fig. S9. Impact of PI3K/Akt/mTOR inhibition on Th cell differentiation *in vitro*. Naïve $CD4^+$ T cells, isolated from indicated mouse lines were cultured for 4 days in the presence of inhibitors (a) Th17 differentiation in the presence of IC-87114 (2 and 10 nM), LY294002 (2 and 10 nM), and CAL-101 (1 and 10 nM). (b) Th17, Treg, and Th1 polarization in the presence of rapamycin (5 and 25 nM). All error bars indicate SEM. *Significant differences (*P* values <0.05) from control or between two groups analyzed by repeated-measures two-way ANOVA with Bonferroni.