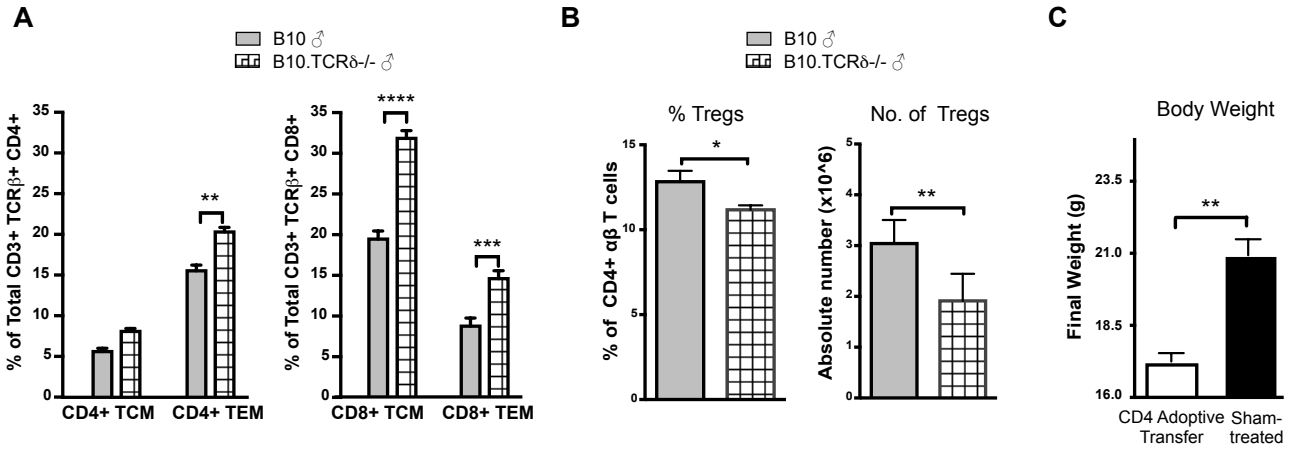
Supplemental Table I

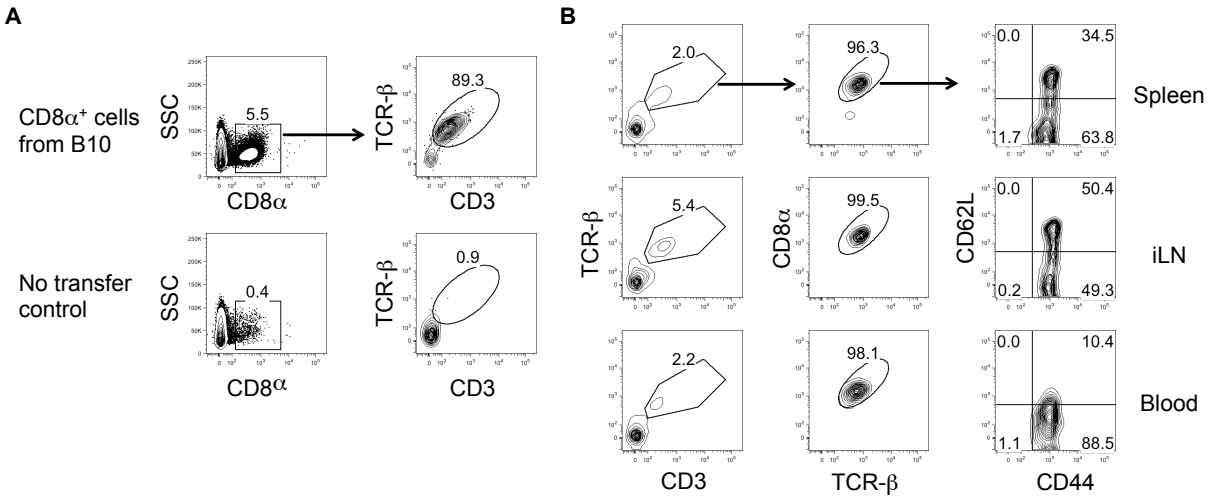
Anti-mouse antibodies and secondary reagents used for flow cytometry and MACS purification

Antibody	**Clone**	**Dyes/Conjugates**	**Source**
Anti-CD3	145-2C11	PE-Cy7	Biolegend
Anti-CD4	GK1.5	FITC/PE/eFluor450	BD Bioscience
Anti-CD8α	53-6.7	PE/Biotin/APC-eFluor780	eBioscience
Anti-CD8β	53-5.8	PE/Biotin	BD Bioscience
Anti-CD11b	M1/70	Percp-Cy™5.5	BD Bioscience
Anti-CD25	PC61	PE/biotin	BD Bioscience
Anti-CD44	1M7	APC	eBioscience
Anti-CD62L	MEL-14	PE	BD Bioscience
Anti-CD122	5H4	PE	eBioscience
Anti-CD124	mIL4R-M1	PE	BD Bioscience
Anti-CD127	A7R34	PE	eBioscience
Anti-Ly6G/C (Gr1)	RB6-8C5	eFluor450	eBioscience
Anti- Ly6G	1A8	FITC/PE	BD Bioscience
Anti-Foxp3		PE-Cy5	Biolegend
Anti-TCR-β	H57-597	PE-Cy5/ APC-eFluor780	eBioscience
Anti-TCR-δ	eBioGL3	APC	eBioscience
Anti-TCR-δ	GL3	FITC/Biotin	Home-made
Anti-TCR-Vγ1	2.11		
Anti-TCR-Vγ4	UC3		
Anti-TNFα	MP6-XT22	PE	BD Bioscience
Anti-IFNγ	XMG1.2	PE/ Percp-Cy™5.5	BD Bioscience
Anti-IL-2	JES6-5H4	PE	BD Bioscience
Anti-IL-6	MP5-20F3	PE	BD Bioscience
Anti-IL-10	JES3-19F1	PE	BD Bioscience
Anti-IL-13	eBio13A	PE	eBioscience
Anti-IL-17A	TC11-18H10	PE	BD Bioscience
Anti-IL-17F	eBio18F10	PE	eBioscience
Streptavidin		FITC	eBioscience
Streptavidin		PE-Cy5	
Streptavidin		APC	
Streptavidin		eFluor450	

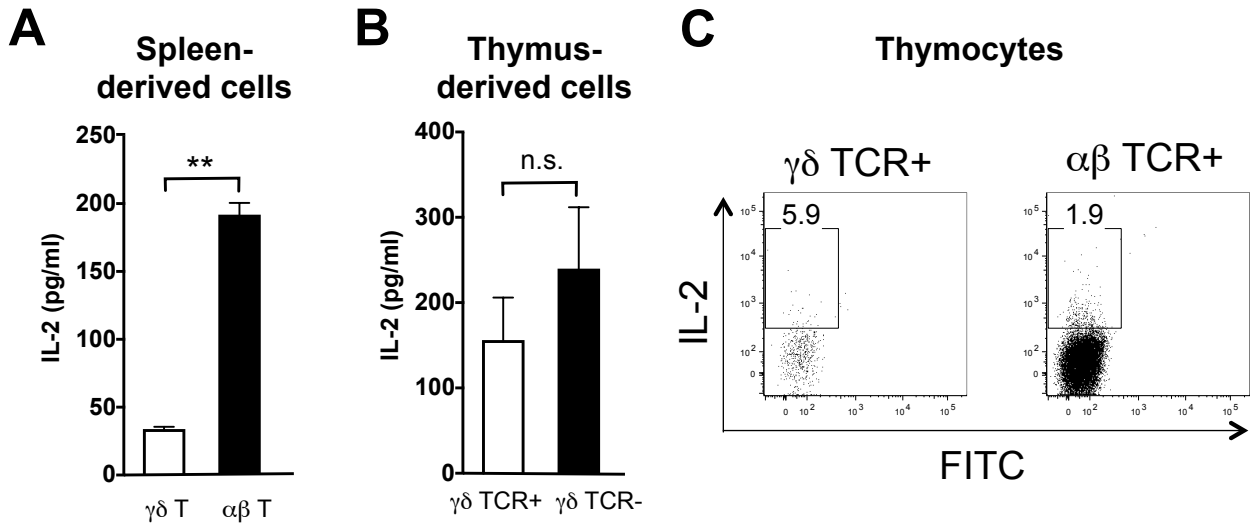
Supplemental Figures



Supp. Fig. 1. A. CD8 memory cells are increased in B10.TCRδ-/- male mice as well. Cells were analyzed as described for Fig. 2 D, except that cells from 3-6 mice per group were tested. Error bars indicates SEM. **B. Tregs are similarly reduced in B10.TCRδ-/- male mice.** Cells were analyzed as described in the legend to Fig. 5A, using 4 B10 male mice and 13 B10.TCRδ-/- male mice. Error bars indicate SEM. **C. CD4+ T cell adoptive transfer results in weight loss.** Final weights of B10.TCRβ/δ–/- females mice shown Fig. 1A, at 18 weeks of age, after being either sham-treated or given an adoptive transfer of CD4+ T cells from the spleens of keratitic B10.TCRδ-/- female donors. Error bars indicate SEM.



Supp. Fig. 2. The composition of CD8α+ cells in B10.TCRβ-δ$^{-/-}$ mice 3 weeks after receiving 2 x 10^5 CD8α+ adoptively transferred cells from B10 mice. A. Flow cytometric analysis of cells from the spleens of these mice showing that virtually all of the CD8α+ cells are CD3+ TCRβ+, and γδ T cells, which would be CD3+ TCRβ-, were not detectable. Note that about 10% of the CD8α+ cells are CD3- and TCRβ-, and likely represent dendritic cells. The flow cytometry profile from the spleen of an untransferred B10.TCRβ-δ$^{-/-}$ mouse was included in the analysis to show that nonspecific staining for CD3 and TCRβ is minimal. **B.** Cells from other tissues of the same mice were analyzed as described in the legend to Fig. 2C. Transferred CD8α+ cells can be found in spleen, lymph node, and blood in the B10.TCRβ-δ$^{-/-}$ recipients, and nearly all display either the TCM (CD44 high, CD62L high) or TEM (CD44-high CD62L low) phenotype.



Supp. Fig. 3. Thymic γδ T cells in B10 mice produce IL-2. A. Spleen cells from 9 weeks old female B10 mice expressing γδ or αβ TCR were purified by positive selection, using biotinylated anti-TCRδ or anti-TCRβ mAbs with streptavidin-complexed MACS beads and columns. Equivalent numbers of each cell type were then cultured in the presence of plate-bound anti-CD3 mAb for 3 days; the supernatants were harvested and tested for IL-2 in an ELISA assay (eBioscience). The mean value obtained from 4 mice is shown; error bars represent SEM. **B.** From the thymi of the same mice as in A, MACs-purified γδ TCR+ and γδ TCR-negative thymocytes (the flow-through cells from the MACS columns) were prepared and similarly analyzed. **C.** Unmanipulated thymocytes from 9 week old female B10 mice were cultured for 6 hours with PMA/ionomycin in the presence of Brefeldin A. After staining with CD3-, TCRβ−, and TCRδ-specific mAbs, cells were fixed, permeabilized with saponin, and stained intracellularly with an IL-2-specific mAb reagent. The panel shows the results of the flow cytometric analysis from a representative mouse. The difference in percent of IL-2+ cells between γδ vs. αβ TCR+ cells was not statistically significant when the means obtained from 4 mice were compared (not shown).