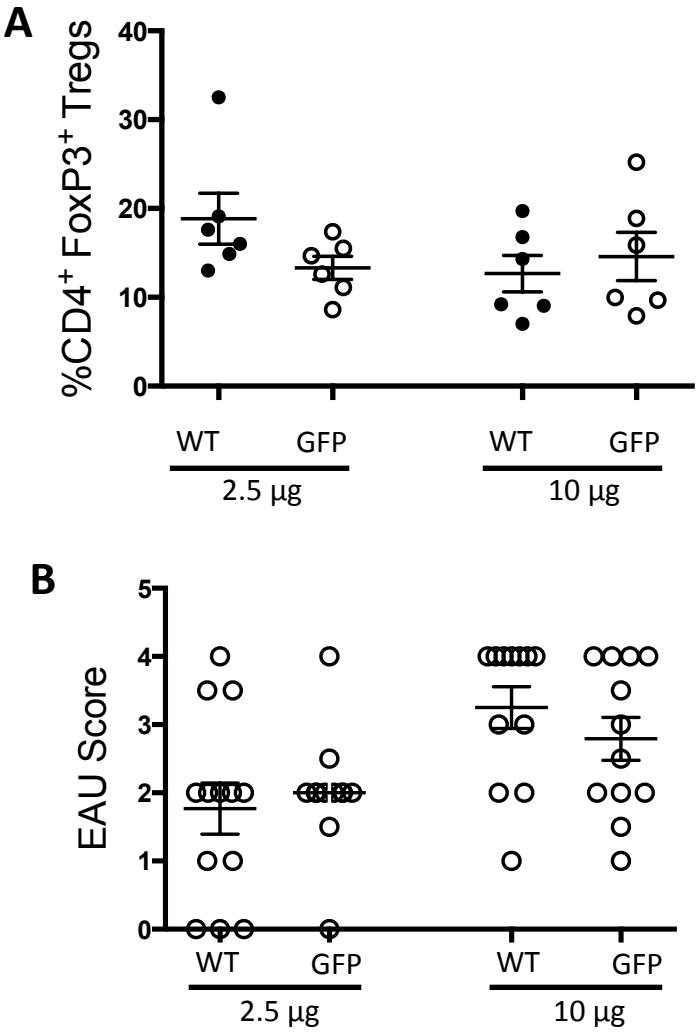

Figure S1. ***WT mice and FoxP3-GFP reporter have similar Treg numbers and develop similar EAU scores in vivo.***

B10.RIII WT and B10.RIII FoxP3-GFP reporter mice (GFP) were immunized with 2.5 or 10 µg of IRBP$_{161\text{-}180}$. Eyes were harvested 21 days later. A) $CD4^+FoxP3^+$ eye cells were detected by flow cytometry. Each symbol represents the combined data of one eye from 2 mice. B) Histology. Each symbol represents one eye of each of the 12 mice per immunization dose (For A and B: Shown is mean +/- SEM. Unpaired t-test= ns for WT vs GFP-reporter mice at all doses).

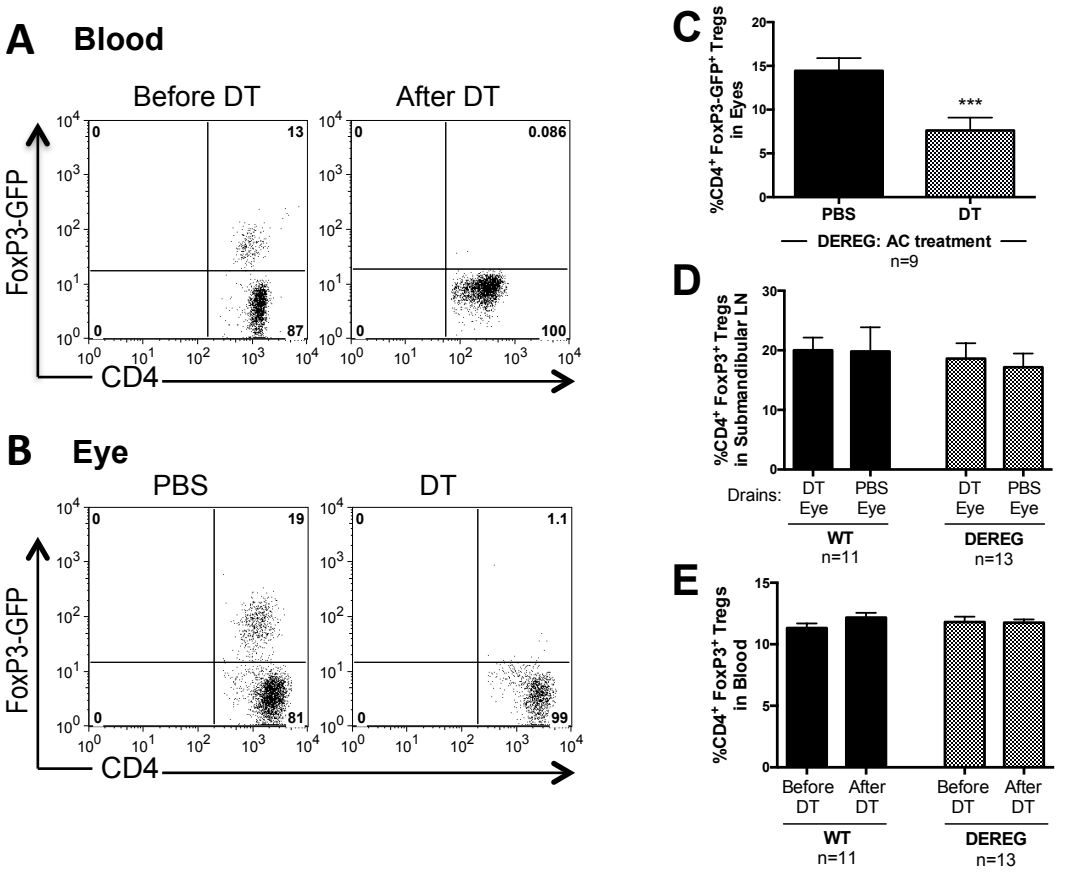

Figure S2. ***Systemic DT injection depletes Tregs in the periphery and in the eye, whereas AC injection of DT depletes Tregs only in the eye.***

The indicated mice (DEREG or WT) were challenged for EAU. **A-B:** DEREG mice were treated with DT systemically. (A) Tregs in blood. DT was injected i.p. at peak disease on 2 successive days. $CD4^+$FoxP3-GFP Tregs were assayed from tail bleeds by flow cytometry before EAU challenge and DT treatment, and again 48 h after the last DT treatment. Shown is one representative mouse. Verification of Treg depletion in blood samples was completed on all experimental mice. (B) Tregs in the eye. $CD4^+$FoxP3-GFP Tregs were assayed (in different mice) 48h after administration of either PBS or DT. Shown is 1 representative experiment of 3. **C–E:** WT or DEREG mice injected with DT into the AC. Contralateral eye received PBS. (C) Eyes. AC delivery of DT depletes Tregs from the eyes of EAU-challenged DEREG mice. $CD4^+$FoxP3-GFP Tregs assayed 36 h after the last treatment (paired t-test: $p<0.0002$ for mouse's PBS-injected eye vs. its own DT-injected eye). (D) Submandibular LNs. The $CD4^+FoxP3^+$ Tregs from the submandibular LN draining the DT-injected eye of each DEREG and WT mouse was compared to that of its own submandibular LN draining the PBS-injected eye 36 h after the last AC treatment (paired t-test=ns). (E) Blood. The $CD4^+FoxP3^+$ Tregs from the blood of each DEREG and each WT mouse 24 h prior to AC treatments was compared to its own blood 36 h after the last DT treatment. (paired t-test=ns). The average value +/- SEM for each group are shown for C, D, and E.

Table S1 Flow cytometric analysis of cells in eyes of individual mice[1]

C57BL/6

	Naive		21 days			3 months		
	FoxP3+	FoxP3-	FoxP3+	%[2]	FoxP3-	FoxP3+	%	FoxP3-
	2	23	5185	10.5	44077	1283	21.6	4666
	8	77	4885	12.1	35427	426	18	1936
	1	9	6579	10.1	58245	521	22.4	1807
	1	1	15044	7.1	197436	727	19.9	2926
	1	1	5218	10	46760	6	7.6	73
	10	8	7688	12.5	54044	1523	20.3	5512
	4	6	7854	12.3	55819			4281
	4	18	13641	19.6	55819			
	3	48						
AV	**4**	**21**	**8,262**	**12**	**68,453**	**748**	**18**	**3,029**

B10.RIII

	Naive		17 days			2 months		
	FoxP3+	FoxP3-	FoxP3+	%	FoxP3-	FoxP3+	%	FoxP3-
	6	38	2962	15.8	15798	21	14.9	121
	4	10	2983	16.6	15032	843	31.7	1818
	3	18	3252	15.3	18020	309	29.7	730
	18	66	4419	14.1	26843	28	26.7	77
	7	12	5860	15.2	32613	191	25.7	553
	6	23	4385	18.6	19202	806	37.8	1328
			3929	18.5	17344			
			2586	17	12609			
			3004	15.2	16736			
			2345	15.5	12796			
AV	**7**	**28**	**3,573**	**16**	**18,699**	**366**	**28**	**771**

[1] Both eyes are combined

[2] percent of total $CD4^+$ cells ($Foxp3^+$ and $Foxp3^-$)

AV = average

Table S2 Infiltrating cells in eyes of individual mice[1]

DT and PBS administered at Peak disease

DEREG MICE		WT Mice	
DT	PBS	DT	PBS
183	115.5	122	286
124	305.5	210.5	205.5
308.5	372	392	163.5
158.5	390	40.5	241.5
50	19	281	260
364	191.7	153	48
452	600	98	161
95.5	472.7	169	255.5
455.5	187.5	52	92.5
374.5	20	80.5	96
233.5	145.5	293.5	336.5
403	23	216.5	266.5
915	724.5	108	86.5
231.5	160	182.5	275.5
434	161.5	57.5	35
278.5	96	409	68
250.5	20.5	480.5	184.5
783.5	318	96	103.5
479	152.5	387.5	173.5
324.5	242	210	116.5
147.5	123	248.5	240
475	134	381.5	250.5
168	97.5	303	350
981.5	233.5	185	202.5
50	0	101.5	159.5
260.5	193.5	355.5	301.5
183.5	123.5	107	109
97	71	175.5	496.5
45.5	27	152	206.5
99	223.5	94.5	646
485.5	326	252	417.5
211	12	504	280.5
106.5	102	165.5	355
114.5	31	217.5	247
266.5	144.5	37	12
323	97.5	174	193
263.5	325	371.5	669.5
136	127.5	415	249.5
276.5	163.7	34	21
164	187	44.5	44.5
525.5	353.5	526.5	356.5
482.5	352	184	127.3
		42.5	58
		299	160
		213.5	135.5
		41.5	79.5
		418.3	110.5
		421	159
		231	81.5
Sum	12761	8166	10736	10175
Mean	303.8	194.4	219.1	207.6

DT and PBS administered after Resolution

DEREG MICE		WT Mice	
DT	PBS	DT	PBS
385.5	38.5	196.5	114
927	77	24	23.5
134.5	115.5	35.5	43
147.5	175.5	152.5	316.5
375.5	136	221	34.5
205.5	174.5	20	119.5
587	216.5	94.5	23.5
365	62.5	130.5	24.5
214	26.5	347.5	124.3
472	282.5	123	24
528	72.5	148	67
299.5	28.5	286.5	267.5
79.5	15.5	212	240
727.5	67	202.5	285.5
316	5.5	117	130
560	412.5	29	25.5
298.5	42.5	25.5	47.5
487	126	11.5	6.5
13	1.5	184.5	105
104	72.5	27	34.5
136.5	59.5	33.5	1
43.5	20	157.7	148.5
23.5	119.5	121	313.5
392.5	41.5	296.5	367.5
		37.5	118
		56	226
		91.5	194
Sum	7823	2390	3382	3425
Mean	325.9	99.56	125.3	126.8

[1]Infiltrating leukocytes in hematoxylin-eosin stained histological sections cut through the pupillary-optic nerve plane were counted under the microscope in a masked fashion. Counts from entire histological sections from each eye were averaged.