

Supplemental Table 1: Reagents used in this study.

Antibody	Clone	Application	Source	Catalog #
rabbit anti-OVA	polyclonal	IB, IP, IF, ELISA	MP Biochemical	0855304
mouse anti-OVA IgG1	OVA-14	IF, ELISA	Sigma	A6075
HRP-conjugated goat anti-rabbit IgG	polyclonal	IP	Southern Biotechnology	4049-05
rabbit anti-mouse/human LDH3	polyclonal	IP, IF, <i>in vivo</i>	gift from Dr. E. Goldberg	--
rabbit anti-mouse/human Zan D3p18	polyclonal	IP, IF, <i>in vivo</i>	gift from Dr. D.M. Hardy	--
rabbit anti-pig Zan	polyclonal	IP	gift from Dr. D.M. Hardy	--
rat anti-mouse CD25	PC61	<i>in vivo</i>	Lymphocyte Culture Center at UVA	--
biotinylated goat anti-rabbit IgG	polyclonal	IF	Southern Biotechnology	4050-08
rat anti-mouse F4/80 IgG	C1:A3-1	IF, FC	Cedarlane	CL8940
biotinylated rabbit anti-rat IgG	polyclonal	IF	Vector Laboratories	BA-4001
biotinylated mouse anti-rat IgG	polyclonal	IF	Jackson Immunoresearch Laboratories	212-066-102
goat anti-rabbit IgG-FITC	polyclonal	IF	Southern Biotechnology	4050-02
goat anti-FITC	polyclonal	IF	Vector Laboratories	SP-0601
rabbit anti-goat FITC	polyclonal	IF	Vector Laboratories	F1-5000
goat anti-rat IgG Biotin	polyclonal	IF	Vector Laboratories	BA-9400
chicken anti-GFP	polyclonal	IF	Abcam	ab13970
goat anti-chicken Alexa Fluor488	polyclonal	IF	Invitrogen	A-11039
chicken anti-goat Alexa Fluor488	polyclonal	IF	Invitrogen	A-21467
chicken anti-rabbit Alexa Fluor488	polyclonal	IF	Invitrogen	A-21441
chicken anti-mouse Alexa Fluor488	polyclonal	IF	Invitrogen	A-21200
chicken anti-rabbit Alexa Fluor594	polyclonal	IF	Invitrogen	A-21442
neutralite avidin-Texas Red	n/a	IF	Southern Biotechnology	7200-07
goat anti-mouse IgG-FITC	polyclonal	IF	Jackson Immunoresearch Laboratories	115-095-003
goat anti-mouse IgG1-FITC	polyclonal	IF	Southern Biotechnology	1070-02
goat anti-mouse IgG2a-FITC	polyclonal	IF	Southern Biotechnology	1080-02
goat anti-mouse C3-FITC	polyclonal	IF	Southern Biotechnology	1090-02
rat anti-mouse occludin	monoclonal 1	IF	gift from Drs. T. Miyamoto & S. Tsukita	--
anti-mouse MHCII	M5/114.7	IF	hybridoma supernatant	--
goat anti-human tACE	polyclonal	IF	Santa Cruz Biotechnology	sc-12187
HRP-conjugated goat anti-mouse IgG	polyclonal	ELISA	Southern Biotechnology	1030-05
rat anti-IFN- $\gamma$	R4-6A2	ELISA	BD Pharmingen	551216
biotinylated rat anti-IFN- $\gamma$	XMG1.2	ELISA	BD Pharmingen	554410
rabbit anti-Claudin-11	polyclonal	IB	Invitrogen	36-4500
rabbit anti-occludin	polyclonal	IB	Invitrogen	71-1500
rabbit anti-ZO-1	polyclonal	IB	Invitrogen	61-7300
rabbit anti- $\alpha$ -catenin	polyclonal	IB	Santa Cruz Biotechnology	sc-7894
rabbit anti- $\beta$ -catenin	polyclonal	IB	Invitrogen	71-2700
goat anti-actin	polyclonal	IB	Santa Cruz Biotechnology	sc-1616
CD4	RM4-5	FC	Ebioscience	47-0042
CD45	30-F11	FC	Ebioscience	17-0451
CD8	53-6.7	FC	Ebioscience	51-008
CD11b	M1/70	FC	Ebioscience	47-0112
DO11.10 TCR	KJ126	FC	Ebioscience	17-9451
CD69	H1.2F3	FC	BD Pharmingen	553235
iNos	CXNFT	FC	Ebioscience	17-5920

MHCII	25-9-17, 10-3.6	FC	BD Pharmingen	553605, 553540
IL-4R $\alpha$	mIL4R-M1	FC	BD Pharmingen	552509
CD206	MR5D3	FC	BD Pharmingen	565250
Foxp3	FJK-16s	FC	eBioscience	17-5773-82
anti-Fc $\gamma$ RII/III	2.4G2	FC	hybridoma supernatant	--

\*IF=immunofluorescence, IP=immunoperoxidase, IB=immunoblot, FC=flow cytometry

Supplemental Table 2: Detailed Procedures for IF and IP studies.

Figure	Purpose (method)	Tissue section	Fixative	1 <sup>st</sup> Ab (dilution)	2 <sup>nd</sup> Ab (dilution)	Amplification	Neutralize Avidin-Texas Red (1:500)	3 <sup>rd</sup> Ab (dilution)	4th Ab (dilution)	Amplification	Neutralize Avidin-Texas Red (1:500)
1B	MGCA & Sertoli cells (IF)	GFP-Sex testes	Bouin	rabbit anti-LDH3 (1:400)	biotinylated goat anti-rabbit IgG (1:200)	-	yes	chicken anti-GFP (1:500)	goat anti-chicken alexafluor488 (1:500)	-	-
1B inset	Occludin & Sertoli cells (IF)	GFP-Sex testes	Acetone/ Ethanol	rat anti-occludin (1:200)	biotinylated goat anti-rat IgG (1:500)	TSA Biotin Amplification Kit	yes	chicken anti-GFP (1:500)	donkey anti-chicken alexafluor488 (1:500)	-	-
2A-D; 3F	Testis Immune Complexes (IF)	WT mouse testes; <i>Ldh3</i> null testes; OVA-Hi & OVA-Lo testes	Acetone/ Ethanol	rabbit anti-LDH3 (1:400), rabbit anti-Zan D3p18 (1:200), rabbit anti-OVA (1:500)	FITC goat anti-rabbit IgG (1:200)	TSA FITC Amplification Kit	-	rat anti-occludin (1:200)	biotinylated goat anti-rat IgG (1:500)	TSA Biotin Amplification Kit	yes
3B; 7A; 7E; 7G	OVA (IP)	OVA-Hi & OVA-Lo testes	Bouin	rabbit anti-OVA (1:500)	biotinylated goat anti-rabbit IgG (1:200)	ABC kit and osmication	-	-	-	-	-
3E	Injected FITC in Seminiferous Tubules (IF)	OVA-Hi testes	Acetone/ Ethanol	rat anti-occludin	biotinylated goat anti-rat IgG (1:500)	TSA Biotin Amplification Kit	yes	-	-	-	-
3G	Testis Immune Complex & MHC class II (IF)	OVA-Hi testes (injected with rabbit anti-OVA)	Acetone/ Ethanol	rat anti-mouse MHCII (1:200)	FITC rabbit anti-rat (1:400)	TSA FITC Amplification Kit	-	biotinylated goat anti-rabbit IgG (1:200)	-	-	yes
6B	Zan & F4/80 (IF)	Control & DERE G testes	Acetone/ Ethanol	biotinylated rat anti-mouse F4/80 (1:300)	-	TSA Biotin Amplification Kit	yes	rabbit anti-Zan D3p18 (1:200)	FITC goat anti-rabbit IgG (1:200)	-	-
6D	Endogenous Immune Complexes & Occludin (IF)	DERE G testes	Acetone/ Ethanol	rat anti-occludin (1:200)	biotinylated goat anti-rat IgG (1:500)	TSA Biotin Amplification Kit	yes	FITC goat anti-mouse IgG (1:100)	-	-	-
6E	Detect Biotin in Seminiferous Tubules (IF)	Control & DERE G testes	Acetone/ Ethanol	-	-	-	yes	-	-	-	-
6G	Rabbit LDH3 Immune Complexes & Endogenous mouse immune Complexes (IF)	DERE G testes	Acetone/ Ethanol	FITC goat anti-mouse IgG (1:100)	biotinylated goat anti-rabbit IgG (1:200)	-	yes	-	-	-	-
7C	OVA & tACE (IF)	OVA-Hi testes	Bouin	mouse anti-OVA (1:200)	chicken anti-mouse alexafluor488 (1:500)	-	-	goat anti-tACE (1:100)	biotinylated rabbit anti-goat (1:200)	-	yes
7D; 7F	OVA & GFP (IF)	GFP-Sex x OVA-Hi testes	Bouin	rabbit anti-OVA (1:500)	biotinylated goat anti-rabbit IgG (1:200)	-	yes	chicken anti-GFP (1:500)	donkey anti-chicken alexafluor488 (1:500)	-	-
8A left	LDH3 (IP)	BALB/c testes	Bouin	rabbit anti-LDH3 (1:200)	biotinylated goat anti-rabbit IgG (1:200)	ABC kit and osmication	-	-	-	-	-
8A right	Zan (IP)	BALB/c testes	Formalin	rabbit anti-Zan D3p18 (1:400)	-	Envision dual link and 3,3'-diaminobenzidine	-	-	-	-	-

						tetrahydrochlori de chromagen (Dako)					
8B	OVA & LDH3 (IF)	OVA- Hi testes	Bouin	mouse anti-OVA	FITC goat anti-mouse IgG (1:100)	-	-	rabbit anti- LDH3 (1:200)	biotinylated goat anti- rabbit IgG (1:400)	-	yes
8C	OVA & Zan (IF)	OVA- Hi testes	Bouin	mouse anti-OVA	FITC goat anti-mouse IgG (1:100)	-	-	rabbit anti-Zan D3p18 (1:200)	biotinylated goat anti- rabbit IgG (1:400)	-	yes
8D	LDH3 & tACE (IF)	BALB/ c testes	Bouin	rabbit anti- LDH3 (1:200)	chicken anti-rabbit alexafluor5 94 (1:500)	-	-	goat anti- tACE (1:100)	chicken anti-goat alexafluor4 88 (1:500)	-	-
8E	Zan & tACE (IF)	BALB/ c testes	Bouin	rabbit anti-Zan D3p18 (1:200)	chicken anti-rabbit alexafluor5 94 (1:500)	-	-	goat anti- tACE (1:100)	chicken anti-goat alexafluor4 88 (1:500)	-	-