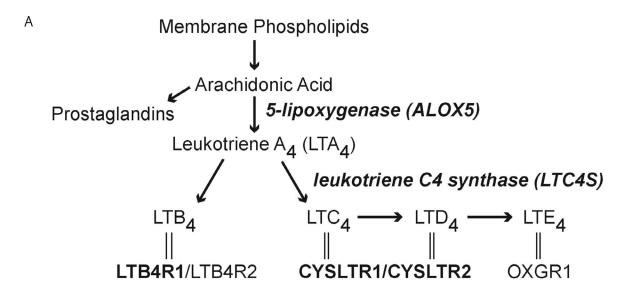
SUPPLEMENTAL MATERIAL

von Moltke et al., https://doi.org/10.1084/jem.20161274



В					С		
Flow Cytometry Antibodies					qRT-PCR Primers		
Target	Conjugation	Source	Clone	Dilution	Target	Forward Primer (5' -> 3')	Reverse Primer (5' -> 3')
CD3	Pacific Blue	Biolegend	17A2	1:100	Cysltr1	TTGAGCCTCCACAGAACAATC	TTCCTACGACTTGGCATGTTTT
CD19	Pacific Blue	Biolegend	6D5	1:100	0.40	T000171110177017070001	
CD11B	Pacific Blue	Biolegend	M1/70	1:100	Cysltr2	TGGGATAAAGATTCATGTGGGGA	AGCCCTTAATCGAGCTTTGAAA
CD11B	BUV-395	BD Biosciences	M1/70	1:500	P2yr12	CCCTGTGCGTCAGAGACTAC	CAAGCTGTTCGTGATGAGCC
CD5	BV-421	Biolegend	53-7.3	1:500	Ltb4r1	ATGGCTGCAAACACTACATCTC	GACCGTGCGTTTCTGCATC
CD5	PE-Cy7	eBioscience	53-7.3	1:500	Ltb4r2	ATGTCTGTCTGCTACCGTCC	AGCTCCATACTACGAAGCCAT
NK1.1	Pacific Blue	Biolegend	PK136	1:100	Overed		GCTGGTCAGATACAGCAAGTC
CD49B	Pacific Blue	Biolegend	DX5	1:100	Oxgr1	CAGCCAGTGATTCGGATTTCC	GCTGGTCAGATACAGCAAGTC
CD8	Pacific Blue	Blolegend	53-6.7	1:100	Rps17	CGCCATTATCCCCAGCAAG	TGTCGGGATCCACCTCAATG
THY1.2	FITC	BD Biosciences	53-2.1	1:500	II33r	AGGCTCTCACTTCTTGGCTGA	GGGACACTCCTTACTTGGTTGT
THY1.2	BV-605	Biolegend	53-2.1	1:500	115	GCAATGAGACGATGAGGCTTC	GCCCCTGAAAGATTTCTCCAATG
CD4	PerCP/Cy5.5	eBioscience	RM4-5	1:100	Gata3	CTCGGCCATTCGTACATGGAA	GGATACCTCTGCACCGTAGC
CD4	FITC	Biolegend	RM4-5	1:100	Galas	CTCGGCCATTCGTACATGGAA	GGATACCTCTGCACCGTAGC
CD4	BV-711	Biolegend	RM4-5	1:100	Tbet	AGCAAGGACGGCGAATGTT	GGGTGGACATATAAGCGGTTC
SIGLEC-F	AF488	BD Biosciences	E50-2440	1:100	Rorc	GACCCACACCTCACAAATTGA	AGTAGGCCACATTACACTGCT
SIGLEC-F	PE	BD Biosciences	E50-2440	1:100	Nfatc1	GACCCGGAGTTCGACTTCG	TGACACTAGGGGACACATAACTG
GR-1	PerCP/Cy5.5	Biolegend	RB6-8C5	1:500	Nfatc2	CCACCACGAGCTATGAGAAGA	GTCAGCGTTTCGGAGCTTCA
ST2	Biotin	MD Bioproducts	DJ8	1:100	IVIAICE	COACOACGAGOTATGAGAAGA	GTOAGOGTTTOGGAGOTTOA
LY6G	APC-Cy7	Biolegend	1A8	1:500	Nfatc3	GCTCGACTTCAAACTCGTCTT	GATGTGGTAAGCCAAGGGATG
CD11C	PE-Cy7	BD Biosciences	HL3	1:100			
Biotin	Streptavidin-APC	Biolegend	N/A	1:100			
human CD4	PE	eBioscience	RPA-T4	5 µl/test			

Figure S1. LT biosynthesis and antibodies and primers used in this study. (A) Schematic of LT biosynthesis pathway. Mice deficient for genes listed in bold were used in this study. (C) Quantitative RT-PCR (qRT-PCR) primers used in this study.

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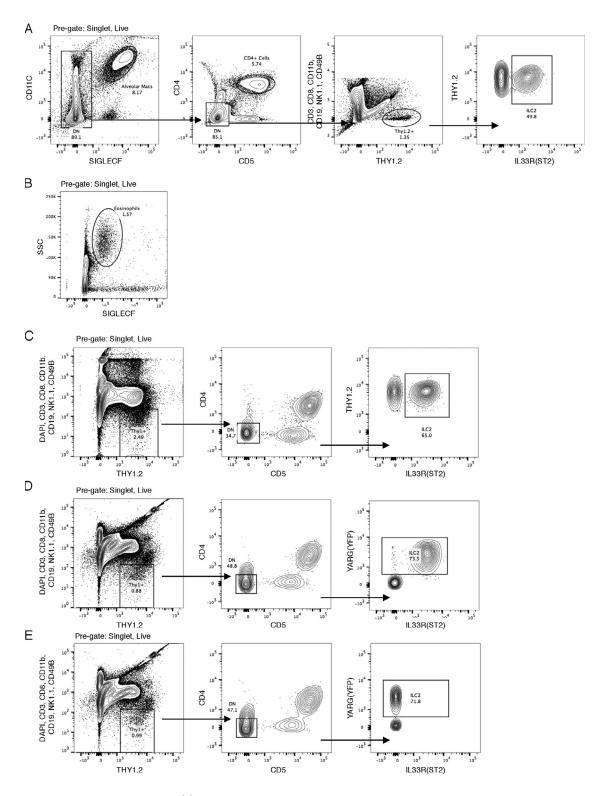


Figure S2. **Gating strategies for cell sorting.** (A) Gating strategy for sorting ILC2 and CD4 cells from the lungs of wild-type mice for quantitative RT-PCR. (B) Gating strategy for sorting eosinophils from blood of wild-type mice for quantitative RT-PCR. (C) Gating strategy for sorting ILC2s from ##II.35mart/Smart mice. (D) Gating strategy for sorting ILC2s from ##Arg17arg/Yarg.;||1135mart/Smart mice with the addition of anti-IL-33R(ST2) antibody. (E) Gating strategy for sorting ILC2s from ##Arg17arg/Yarg.;||1135mart/Smart mice without anti-IL-33R(ST2) antibody. Data are representative of at least three independent experiments. mac, macrophage; SSC, side scatter.

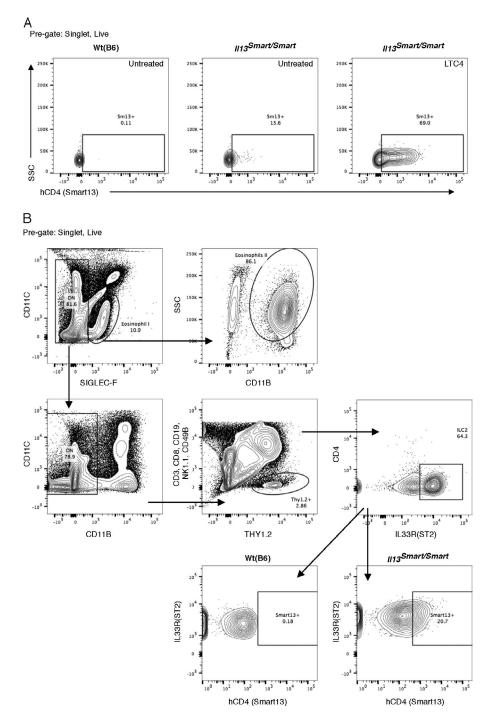


Figure S3. **Flow cytometry gating strategies.** (A) Representative Smart13 reporter gating for ILC2s sorted from the indicated mice and stimulated 6 h in vitro as indicated. (B) Gating strategy for analysis of lung eosinophils, ILC2s, and in vivo Smart13 reporter expression. Data are representative of at least three independent experiments. SSC, side scatter.

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