SUPPLEMENTAL MATERIAL

Yu et al., http://www.jem.org/cgi/content/full/jem.20121846/DC1

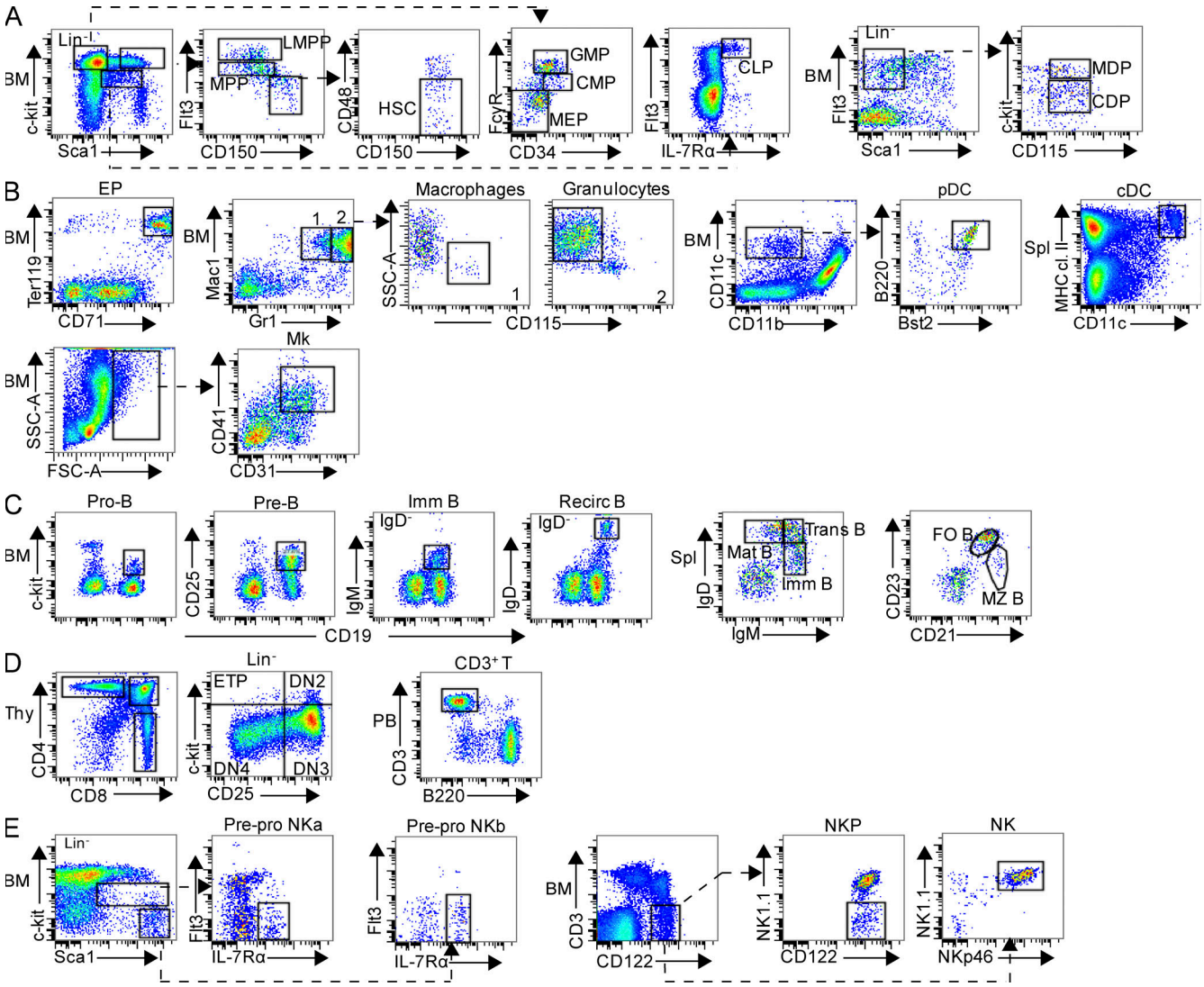

Figure S1. Gating strategies of hematopoietic populations. (A) HSC, MPP, LMPP, CMP, GMP, MEP, CLP, monocyte-dendritic precursor (MDP) and CDP. (B) EPs, macrophages, granulocytes, plasmacytoid dendritic cells (pDC), conventional dendritic cells (cDC), and megakaryocytes. (C) B cell compartment. (D) T cells. (E) NK cells in the BM. In all flow cytometry assays, at least 4 wild-type mice were analyzed for each cell type in independent experiments.

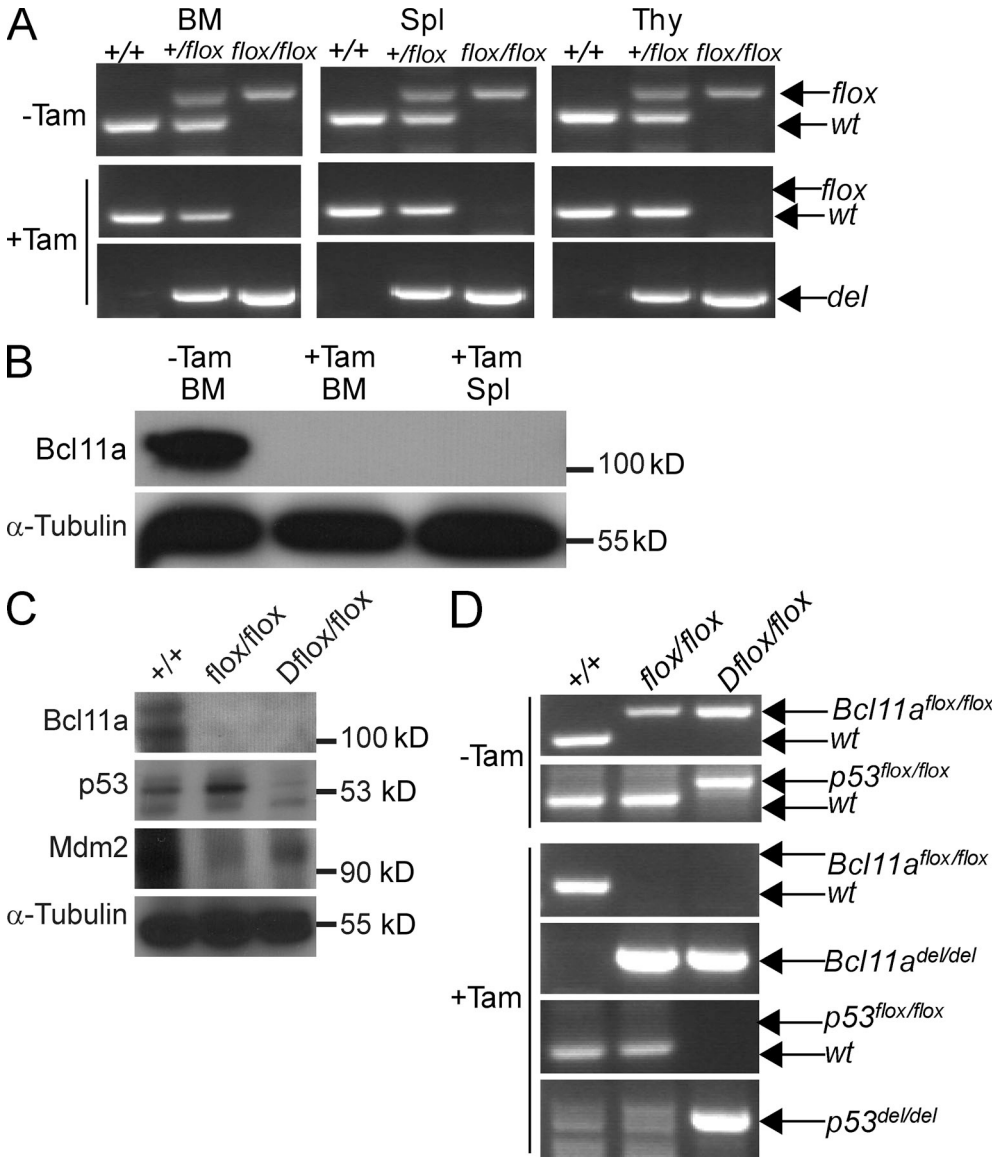

Figure S2. Genotyping strategies. (A) Genotyping cells of the BM, spleen (Spl), and thymus (Thy) for *Bcl11a* deletion (absence of the *flox* allele). wt, wild-type allele; flox, conditional knockout allele; del, deletion allele. (B) Western blotting analysis of BM and Spl cells for Bcl11a protein. α-Tubulin was the loading control. (C) Western blot analysis of Bcl11a, p53, and Mdm2 in cultured BM B cells of indicated genotypes. Loading control, α-tubulin. (D) Genotyping of cells for *Bcl11a* and *p53* deletion. $Bcl11a^{flox/flox}$, *Bcl11a* conditional knockout allele; $p53^{flox/flox}$, *p53* conditional knockout allele; $Bcl11a^{del/del}$: Bcl11a deletion allele. $p53^{del/del}$: *p53* deletion allele.

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Table S1. Hematopoietic population definitions

Tissue	Population	Makers	Lineage
Bone marrow	HSC	$Lin^-Sca1^+c\text{-}kit^+$ $Flt3^-CD48^-CD150^+$	B220,CD3ε,CD4,
	MPP	$Lin^-Sca1^+c\text{-}kit^+$ $Flt3^{inter}CD150^-$	CD8α,NK1.1,
	LMPP	$Lin^-Sca1^+c\text{-}kit^+$ $Flt3^+$	CD11b,Ly6G,
	CLP	Lin^- $Flt3^+IL\text{-}7R\alpha^+sca1^{low}c\text{-}kit^{low}$	Ter119
	Pre-pro NKa	$Lin^-Sca1^+c\text{-}kit^{int}Flt3^-IL\text{-}7R\alpha^+$	
	Pre-pro NKb	$Lin^-Sca1^+c\text{-}kit^-Flt3^-IL\text{-}7R\alpha^+$	
	CMP	$Lin^-Sca1^-c\text{-}kit^+Fc\gamma R^{int}CD34^{int}$	B220,CD19
	GMP	$Lin^-Sca1^-c\text{-}kit^+$ $Fc\gamma R^{hi}CD34^{hi}$	CD3ε,CD4,CD8α,
	MEP	$Lin^-Sca1^-c\text{-}kit^+$ $Fc\gamma R^{low}CD34^{low}$	NK1.1,Ly6G,
	MDP	$Lin^-Sca1^-c\text{-}kit^{hi}Flt3^+CD115^+$	IgM,IL-7Rα,
	CDP	$Lin^-Sca1^-c\text{-}kit^{low}Flt3^+CD115^+$	Ter119
	Pro-B	$B220^+CD19^+CD43^+c\text{-}Kit^+IgM^-$	
	Pre-B	$B220^+CD19^+CD43^-CD25^+IgM^-$	
	Imm B	$B220^+CD19^+IgM^{hi}IgD^-$	
	Recirc B	$B220^+CD19^+IgD^+$	
	NKP	$CD122^+NK1.1^-CD3^-$	
	NK	$NK1.1^+CD122^+NKp46^+CD3^-$	
	Macrophages	$Mac1^+Gr1^{low}CD115^+$	
	Granulocytes	$Mac1^+Gr1^{hi}CD115^-$	
	EP	$CD11b^-Ly6G^-CD3^-B220^-Ter119^+CD71^+$	
	pDC	$CD11b^-CD11c^{low}B220^+Bst2^+$	
	cDC	$CD11C^{hi}$MHC cl.II$^+$	
	Megakaryocytes	$FSC^{hi}CD41^+CD31^+$	
Thymus	ETP	$Lin^-c\text{-}kit^{hi}CD25^-$	B220,CD19,CD3ε,
	DN2	$Lin\text{-}c\text{-}kit^{hi}CD25^+$	CD8α,TCRβ,TCRγδ,
	DN3	$Lin^-c\text{-}kit^-CD25^+$	NK1.1,CD11c,CD11b
	DN4	$Lin^-c\text{-}kit^-CD25^-$	,Ly-6G,Ter119
Spleen	Imm B	$CD19^+IgM^{hi}IgD^-$	
	Trans B	$CD19^+IgM^{hi}IgD^{hi}$	
	Mat B	$CD19^+IgM^{low}IgD^-$	
	MZ B	$CD21^{hi}CD23^{low}$	
	FO B	$CD21^{int}CD23^{hi}$	
	NK	$NK1.1^+Ter119^-Gr1^-Mac1^-B220^-CD3^-$	
	$CD3^+$ T	$Ter119^-Gr1\text{-}Mac1^-B220^-CD3^+$	
Peripheral blood	B	$Ter119^-Gr1\text{-}Mac1^-B220^+CD3^-$	
	T	$Ter119^-Gr1\text{-}Mac^-B220^-CD3^+$	
	NK	$NK1.1^+Ter119^-Gr1^-Mac^-B220^-CD3^-$	
	GM	$Ter119^-Mac1^+Gr1^+CD3^-B220^-$	

Table S2. Antibodies in this study

Epitope	Clone	Conjugate	Company
B220	RA3-6B2	Brilliant Violet 421	BioLegend
	RA3-6B2	FITC	eBioscience
	RA3-6B2	PE	eBioscience
	RA3-6B2	APC-Cy7	BioLegend
	RA3-6B2	PerCP-Cy5.5	BioLegend
CD19	RA3-6B2	PE-Cy5.5	BD
	6D5	Brilliant Violet 421	BioLegend
	1D3	FITC	eBioscience
	1D3	PE	BD
	1D3	APC	eBioscience
	1D3	PE-Cy5.5	eBioscience
CD3ε	145-2C11	FITC	eBioscience
	145-2C11	PE	BD
	145-2C11	Alexa Fluor 647	BioLegend
	145-2C11	PE-Cy5.5	BD
CD4	RM4-5	FITC	eBioscience
	RM4-5	PE	BD
	RM4-5	PE-Cy5.5	BD
CD8α	53-6.7	FITC	BioLegend
	53-6.7	PE	BD
	53-6.7	PE-Cy5.5	BD
TCRβ	B20.6	FITC	BD
	H57-597	PE-Cy5.5	BD
DX5	DX5	FITC	BD
	DX5	PE	BD
TCRγδ	GL3	FITC	BD
NK1.1	PK136	FITC	BD
	PK136	PE	eBioscience
	PK136	PerCP-Cy5.5	BD
	PK136	PE-Cy5.5	BioLegend
CD11b	M1/70	FITC	eBioscience
	M1/70	PE	BD
	M1/70	PE-Cy7	eBioscience
	M1/70	PE-Cy5.5	eBioscience
CD11c	N418	FITC	eBioscience
	HL3	PE	BD
	N418	PE-Cy5	eBioscience
	N418	Brilliant Violet 421	BioLegend
Gr1	RB6-8C5	FITC	BD
	RB6-8C5	PE	BioLegend
	RB6-8C5	PE-Cy5.5	eBioscience
Ter119	TER-119	FITC	eBioscience
	TER-119	PE	eBioscience
	TER-119	APC	BioLegend
	TER-119	PE-Cy5.5	BioLegend
IgM	RMM-1	FITC	BioLegend
	RMM-1	PE	BioLegend

	II/41	PE-Cy5	eBioscience
IgD	11-26C.2A	FITC	BD
	11-26C	APC	eBioscience
c-kit	2B8	PerCP-Cy5.5	BioLegend
	2B8	Brilliant Violet 421	BioLegend
Flt3	A2F10	PE	eBioscience
Sca1	D7	APC	eBioscience
	D7	PE-Cy7	BD
	D7	V450	BD
IL-7Rα	SB/199	FITC	BioLegend
	SB/199	PE	BD
	A7R34	APC	BioLegend
	A7R34	Brilliant Violet 421	BioLegend
FcγR	2.4G2	PE-Cy7	BD
CD34	MEC 14.7	Brilliant Violet 421	BioLegend
	RAM34	Alexa Fluor 700	BD
CD150	TC15-12F 12.2	APC	BioLegend
CD48	HM48-1	Alexa Fluor 700	BioLegend
CD71	RI7217	PE	BioLegend
CD25	PC61	APC	BD
Ly6d	49-H4	Alexa Fluor 647	BD
CD115	AFS98	APC	BioLegend
CD43	S7	FITC	BD
	S7	PE	BD
CD317(Bst2)	927	Pacific Blue	BioLegend
MHC cl.II	M5/114.15.2	PerCP-Cy5.5	BioLegend
CD41	MWReg30	PE	BioLegend
	MWReg30	APC	eBioscience
CD31	MEC13.3	APC	BioLegend
CD21	eBio8D9	PE	eBioscience
	4E3	eFluor450	eBioscience
CD23	B3B4	PE	BD
CD45.1	A20	APC-eFluor 780	eBioscience
CD122	TM-B1	eFluor450	eBioscience

Table S3. Primers used in this study

Genotyping primers		
Genotyping PCR primers	Primer sequence (5′-3′)	Size of PCR products (bp))
Bcl11a-cko-FW	TAGCTCCTGCTAGCCAGGTTTCTT	377(wild type)
Bcl11a-cko-RV	CGAGGCTTGCAGAAACAGAAAGAT	470(*flox*)
Bcl11a-cko-DEL	CTCGAAGGGAGGTTTCGGTATTGTG	700(*del*)
Bcl11a-eGFP-5′-FW	GAGACAAGAACAGGTGCAAGAGTGGATT	6746(targeted)
Bcl11a-eGFP-5′-RV	GGGTTATTGAATATGATCGGAATTGGGC	
Bcl11a-eGFP-3′-FW	GAAAGAACCAGCTGGGGCTCGACTAGAG	4507(targeted)
Bcl11a-eGFP-3′-RV	CAGCGAGGTCCCCTTTCTCACTAAAAAT	

Primers for ChIP assay qPCR	
PCR primers	Primer sequence (5′-3′)
Mdm2-Fwd	GCCAGCGTAGCCTAGGAG
Mdm2-Rev	AAGAGGGAATGATCGGGACT
Mdm4-Fwd	CAGGGTCCGCTCTATGGTT
Mdm4-Rev	ACCCCAACACCACACCTTAC
Cdkn1a-Fwd	GGGCCAGAGCTAAGAGCAC
Cdkn1a-Rev	CACGCACGTACACAGACACA
Bcl2-Fwd	GGGGACAAGCTGTTGTAGGA
Bcl2-Rev	AGAGGGGACAGATAGCGATG
Bcl2l1-Fwd	GCTGTGCAGGGAGGAGATAA
Bcl2l1-Rev	CTCTGGGAGTAGGTGGTTGC
Pax5-Fwd	GTGTGTGCATGAGCATGTGA
Pax5-Rev	CCATTCCATGTGCAGAGATG
Tcf3-Fwd	GGGACAGAGTGAGGACAGTCA
Tcf3-Rev	CCCCATCTACTCTGTCACCA

qRT-PCR primers			
Gene name	Forward primer (5′-3′)	Reverse primer (5′-3′)	Company
Bcl11a	AOD Probe	Mm01225235_m1	Applied Biosystems
Sfpi1	AOD Probe	Mm00488142_m1	Applied Biosystems
Ikzf1	AOD Probe	Mm01187882_m1	Applied Biosystems
Tcf3	AOD Probe	Mm01175591_m1	Applied Biosystems
Ebf1	AOD Probe	Mm00395519_m1	Applied Biosystems
Pax5	AOD Probe	Mm00435501_m1	Applied Biosystems
Cd79a	AOD Probe	Mm00432423_m1	Applied Biosystems
Foxo1	AOD Probe	Mm00490672_m1	Applied Biosystems
Il7r	AOD Probe	Mm00434295_m1	Applied Biosystems
Stat5a	AOD Probe	Mm03053818_s1	Applied Biosystems
Notch1	AOD Probe	Mm00435249_m1	Applied Biosystems
Bcl11b	AOD Probe	Mm00480516_m1	Applied Biosystems
Cebpa	AOD Probe	Mm00514283_s1	Applied Biosystems
Pbx1	AOD Probe	Mm04207617_m1	Applied Biosystems
Runx1	AOD Probe	Mm01213405_m1	Applied Biosystems
Mef2c	AOD Probe	Mm01340842_m1	Applied Biosystems
Ccr9	AOD Probe	Mm01718955_g1	Applied Biosystems
Flt3	AOD Probe	Mm00439016_m1	Applied Biosystems
Foxp1	AOD Probe	Mm00474848_m1	Applied Biosystems
Mpo	AOD Probe	Mm01298424_m1	Applied Biosystems
Csf3r	AOD Probe	Mm00432735_m1	Applied Biosystems

Table S4. Lymphoid genes enriched for clustering from microarray data

Sfpi1	*Foxo3a*	*Tcf7*	*Scd1*
Ikzf1	*Pten*	*Mef2c*	*Car13*
TCF3	*Dntt*	*Gfi1*	*Cplx2*
EBF1	*Ets1*	*Evi1*	*E2f2*
Pax5	*Lef1*	*Bach2*	*Ikzf3*
Foxo1	*Syk*	*Hes1*	*Cbx2*
Cd79a	*Runx1*	*Xbp1*	*Hes5*
Cd79b	*Runx2*	*Igh-6*	*Pold4*
Vpreb1	*Runx3*	*Igll 1*	*Zfpm1*
Vpreb2	*Id2*	*Klf4*	*Dtx1*
Vpreb3	*Id3*	*C-kit*	*Bcar3*
Cd19	*Sox4*	*Cebpb*	*Nedd9*
Erg	*Stat3*	*Ccnd3*	*Sit1*
Blk	*IL-7Rα*	*Bcl7a*	*Btg2*
Ccr7	*Stat5a*	*Bcl7b*	*Tox*
Ccr9	*Stat5b*	*Cd93*	*Tmem66*
Cxcr4	*Sirt1*	*Flt3*	*CD22*
Pou2f1	*Sirt3*	*Foxo6*	*Evx1*
Pou2af1	*Gata3*	*Ccn3*	*Ncf4*
Rag1	*Notch1*	*Bank1*	*Tbp*
Rag2	*Notch2*	*Tcf4*	*Zap70*
Irf4	*Notch3*	*Bst1*	*Pde2*
Irf8	*Stat1*	*Klf7*	*Ccr2*
Blnk	*Foxp1*	*Clgn*	*Parp1*
Ezh2	*Bcl6*	*Car2*	*Hdac7*