

SUPPLEMENTARY INFORMATION

TABLE S1. Sequences of primers used.

Figure S1. RUNX binding sites are conserved at *NCR1* promoter. *NCR1* proximal upstream regions in 10 mammals were aligned using EBI Clustal2W. Bases identical in at least 8 species are highlighted in yellow. TESS sites that are found in multiple species are boxed. Note only RUNX and ETS sites are highly conserved. The distal RUNX site in mouse and rat are found in the antisense direction and offset by a few base pairs, suggesting convergent evolution. The tissue specific and essential regulatory sequences are marked by solid and dashed lines under the sequence, respectively.

Figure S2. Promoter constructs transfected into KY-2 mouse NK cell line. 2.5×10^5 KY-2 cells were transfected using Lipofectamine 2000 (Invitrogen) according to manufacturer instructions. Transfection mix consisted of 1 μ g vector, 0.1 μ g pRL-TK, and 2 μ g Lipofectamine 2000. Cells were incubated for 24 hours before assaying using the Dual-Luciferase Reporter Assay System (Promega) according to the manufacturer's instructions. Firefly luciferase activity was normalized relative to Renilla luciferase activity for each transfection and calculated as fold increase over pGL4.10-BASIC (pGL4B).

Figure S3. *Ncr1*, *Runx1*, *Runx3* transcripts in mouse. (A) RT-PCR detection in mouse cell lines with *Actb* as internal control. Total RNA from indicated cell lines was used for reverse transcriptase reactions. (B) Microarray of *Runx1* and *Runx3* expression. Data accessed through BioGPS (51). Mouse data from GeneAtlas MOE430 probe set 1427650_a_at for *Runx1* and 1440275_at for *Runx3* (53). Only relevant hematopoietic cell types are shown.

Table S1

Primer set name	Forward Primer	Reverse Primer
hNCR1-RT-PCR	5'caccctctcggttcatcc	5'gagattctgggcagtgtg
hACTB-RT-PCR	5'aaggagatcactgccctggc	5'ccacatctgctggaaggtgg
5'95-pGL4B	5'aaggtaccgctggtgctcaccacc	5'aagctagctgctcagattctgccgg
5'200-pGL4B	5'aaggtaccgtgtcagagggaccacgg	Same as above
5'270-pGL4B	5'aaggtaccgagaagtgaccagaaatgc	Same as above
5'395-pGL4B	5'aaggtaccctgttccagtatctcactg	Same as above
3'276-pGL4B	Same as above	5'aagctagcactgctttcatcagaacg
3'196-pGL4B	Same as above	5'aagctagcatctgtggtctagccagc
3'105-pGL4B	Same as above	5'aagctagctaccagagctgactgtg
hPU.1-RT-PCR	5'ccaacgcacagattatcc	5'aagctctgaactcgtctgtg
hCJUN-RT-PCR	5'aaggaagctggagagaatcg	5'tgtttaagctgtgccacctg
hJUNB-RT-PCR	5'cgatctgcacaagatgaacc	5'tgctgaggttggtgtaaacc
hJUND-RT-PCR	5'gcctcatcatccagtccaac	5'tctgcttggtgtaaatcctccag
hCEBPa-RT-PCR	5'tggacaagaacagcaacagag	5'ttctcactggtcagctccag
hMYB-RT-PCR	5'cagcccactgttaacaacgac	5'ctggctgaggacattgac
hMEIS1-RT-PCR	5'ggaggatcaaaatcagacagtg	5'tcctctgaacgagtagatgc
hCP2-RT-PCR	5'ggtgaagagtatttccgtgtgg	5'attctgtctccaggtcggttc
hE2A-RT-PCR	5'gaagcagcagcagctttg	5'gagaaggaggatgcagatgg
hRUNX1-qPCR	5'tcaggtttgtcggtcgaag	5'gccatccactgtgattttg
hRUNX3-qPCR	5'gttcaacgaccttgccttc	5'gtccacggcactctgatg
hRUNX3dist-RT-PCR	5'acactctgccgaccttcat	Same as above
hRUNX3prox-RT-PCR	5'tattcccgtagaccaagca	Same as above
mNcr1-RT-PCR	5'tggctcttacaacgactatgc	5'agaagaagtaggctcggtaggtg
mRunx1-RT-PCR	5'tagcgagattcaacgacctc	5'atgtaggtggcaactttgtg
mRunx3-RT-PCR	5'ggttcaacgaccttgcattc	5'cggtgtaggttagccacttg
mActb-RT-PCR	5'aaggccaacctgaaaagat	5'gtggtacgaccagaggcatac
distRUNXmut	5'gagttgctggctagacgttagatgtgtcagaggg	5'ccctctgacacatctaactctagccagcaactc
proxRUNXmut	5'gctggtgctcaccacggttctctgtatctatc	5'gatagatacacaggaaaactggtgtagcaccagc
hNCR1promoter-ChIP	5'ctgatgaagcagctcaacgtg	5'gggccaggagatagatacac
h+8intergenic-ChIP	5'tatcttgacaaggctcagc	5'gcttcagcactgaatgatcc
hCD122promoter-ChIP	5'caagcaggtccctctaggtg	5'taacggagtaaggggcttc
mNCR1promoter-ChIP	5'aaatgggtgcagactgagc	5'accattgacctaggactcagg
mIfng-qPCR	5'cacggcacagctcattgaag	5'ccagttcctccagatatccaag
mPrf1-qPCR	5'gtgtcgcattgtacagttttcg	5'tgtgtaagcatgctctgtg
m18S-qPCR	5'gtaaccggtgacccccatt	5'ccatccaatcggtagtagcg
mB2m-qPCR	5'catggtcgtcgtggtgacc	5'aatgtgagcgggtggaactg
mCD43-qPCR	5'agtttcttgaccaccttgg	5'ttctggaagcagtgctgatg
EMSA(enhancer)-WT	5'ggtgctgctag accaca gatgtgtcagag	5'ctctgacacatct gtggt ctagccagcaac
EMSA(enhancer)-MU	5'ggtgctgctag actgta gatgtgtcagag	5'ctctgacacat taactg ctagccagcaac
EMSA(essential)-WT	5'tggtgctcacc accact ctctgtgtatct	5'agatacacagga agtggt gggtgagcacca
EMSA(essential)-MU	5'tggtgctcacc actgtt ctctgtgtatct	5'agatacacagga aaactg gggtgagcacca

*For EMSA oligos, putative RUNX binding sites and mutated sites are shown in bold.

Human	-----CACATTCGTAAAT--TTCTTTT	AATTCCTGTTCCAGTATCTCACTGTGAAACTCCC-----TATGTTTTTATACGATTCTC
Rhesus	-----CAC-TCCTTTAAT--TTCTTTT	CATTCCTGTTCCAGTATCTCACTGTGAAATTCCC-----TGTGTTTTTATACGATTCTC
Armadillo	-----	-----
Cow	-----	-----CA-----CATATTTTTAAAATGACTTT
Horse	GAATAGAAATTTTATTAAAACATTGTAACAAATCTTTAAAGAAGATAGTGTT--ATCATT	CATTCCTGATGAGGTCAATTAATTT-----ATCCA-----AGCATATACATGCTGTACAC
Panda	-----	CATCCTGGTGAAACCAATTCCTG-----ACCCA-----AGCATATGCATACCAAGAG
Elephant	-----	-----
Dog	-----CACAAAGTTTTTAAATGATCTAAC	ATTTCTGCTTGGTACACTACCCTATGCCTTTCCCTGGGCATAAATGTTCCACATTTTCATAATT
Mouse	-----	-----TATTGGG-----GAGTCCCTCAGACCT-
Rat	-----	-----TATTGGG-----AAATCCTCAGACCT-

			C/EBP
Human	AGGGGG-----TTTCCTCTGGGCATGATTGGGCACA-----A	CTTCCCACAGTCAGCTCTGGGTA--CGACCTCCACATTGCAGAAT-----TGAGAAGT	
Rhesus	AGGGGG-----TTTCCTCTGAGCGTCATTAGGCCTG-----A	CTTCTCAGGTCAGCTCTGGGTA--CCACCTCCACATTGCAGAAT-----TGAGAAGC	
Armadillo	-----	-----AT-----TRAGAGAT	
Cow	AAGGGT-----TTTGCTCTGTAC-----A	CTGCCCTATGCCTTCCCTGGGCA--TAAAGTCCCAATTCAAAAT-----TTACCAGT	
Horse	AGGGGAAGCAGGGTTAAACAC-----TTTTTTAAATGATTCTAAGGTTTTTTCTCTGTACA	CTGCCCATGCCTTTTCTGGACA--TAATGTCCACATTTCAAAAT-----TTGGANGT	
Panda	AGGGGAGCCAAGATTAACACATAATTTTTTTAAATGATTCT-	-----TTCTTGGCA--TAATGTCCACATTTCAAAAT-----TTGGANGA	
Elephant	-----	-----CCCTGGGCA--TAATGTCCACATTTCAAAAT-----GTAGAAGT	
Dog	TAGAAG-----TGAACCCCTGATTAGTAAATGAAAC-----A	CCAATAAAAAATAAATTTAAAAAATAAAAAAATAAATAAATAAATAAATACC	
Mouse	-----TTTGCTCTGTGAAT-----	-----CTCCCCATGCC-----TTCCTTGAATGAAT-----TTAAATGT	
Rat	-----TTTGCTCTGTGAAT-----	-----CTCCTCCATGCC-----TTCCTTGAATGGAT-----TTGTAGGT	

	C/EBP		RUNX	
Human	TGACCCAGAAATGCATTTTGGGCTGAG--CAGACAATTGTCAGAGTTGCTGGCTAGA--C	CACAGATGTG-TCAGA-----GGGACCACGGCCTTT---CTGTAA-GCTCAT--G	CACAGATGTG-TCAGA-----GGGACCACGGCCTTT---CTGTAA-GCTCAT--G	
Rhesus	TGACCCAGAAATGCATTTTGGGCTGAG--CAGACAAGTGTTCAGAGGAGCTGGCTAGA--C	CGCAGATGTG-TCAGA-----GGGACCATGGCCTTT---CCATAG-GCTCAT--G	CGCAGATGTG-TCAGA-----GGGACCATGGCCTTT---CCATAG-GCTCAT--G	
Armadillo	TGACTCAGAAA--GAGTGGCGGGCGGA--TAGACGATTTTCAGAGGCTCTCGCTGGA--C	CACAGGGGCGGTCAGAT--TCATGGGCAGCATGTCTTC---CTTCAG-ACCTCATAG	CACAGGGGCGGTCAGAT--TCATGGGCAGCATGTCTTC---CTTCAG-ACCTCATAG	
Cow	TGACCTAGAAAATGAGTTTCTGCCAGAAC--CAGACCATGTCAGAGGCTCTAGTTAGAAAC	CACAGATACACTCAGATGCATCATGGAACTACTCTTTT---CCTTACTGCAT--G	CACAGATACACTCAGATGCATCATGGAACTACTCTTTT---CCTTACTGCAT--G	
Horse	TGACTCATAAATGAGTTTCTAGTGTGAGG--CAGGCAATGTCAGAGGATCTGGTTAGA--C	CGCAGATATGACCAGATGTGTCGTGAGTACCATG---T---TCTTACGCCCAT--G	CGCAGATATGACCAGATGTGTCGTGAGTACCATG---T---TCTTACGCCCAT--G	
Panda	GGACCTGAAATGAGTTTCTAGGTTGGG--CAGACAATTGTCAGAGGATCTGGTTAGA--C	CACAGACAGATCAGATGTGTCATGGGACCATGACCTGT---CPTTATGCCCTAT--G	CACAGACAGATCAGATGTGTCATGGGACCATGACCTGT---CPTTATGCCCTAT--G	
Elephant	GGAAACCAGAAATGGTTTTTCAGGCCAGA--CAAACAACCATTAG-----TTAGA--C	CACACATGTGGTCAGAAGTTCATAGATGCCATGCTCTTTTATCCCTCAGACCCTG--G	CACACATGTGGTCAGAAGTTCATAGATGCCATGCTCTTTTATCCCTCAGACCCTG--G	
Dog	GAACCTGAGATGAGTCTCAGGTGAGG--CAGACAATTGTCAGAGGATCTGGTTAGA--C	CACAGATGATCAGATATGTCATAGGGACCATGATTTGT---CTCAT--G	CACAGATGATCAGATATGTCATAGGGACCATGATTTGT---CTCAT--G	
Mouse	TAACCCAGAAATGGTTGCACTGAG--CAGTTGATGTCAGAAATAATCTTTAGAT-C	CA-AAATGTGGTTAAATGTGTATGTGAACCAAGTTTGTG---CCTGAAGTCTAA--G	CA-AAATGTGGTTAAATGTGTATGTGAACCAAGTTTGTG---CCTGAAGTCTAA--G	
Rat	TAACCCAGAAATCGATTGCACTGAG--CAGTTGATGTCAGAGTAACTTCTAGAT-C	CA-AAATGTGGTTAAATGTGTATGTGAACCAAGCCTTTG---TCTAAAGTCCAA--G	CA-AAATGTGGTTAAATGTGTATGTGAACCAAGCCTTTG---TCTAAAGTCCAA--G	

		RUNX	ETS
Human	GTCAGAGGCGGAGGGG-AGTTGTGAA--CGTTCGTAT---GAAAGCAGTCAACGTGAAAG	CGCTCTGGTGATGGG---CGTGGTGCTCACCCACCACCTCCTGTGATCTATCTCCCTG	CGCTCTGGTGATGGG---CGTGGTGCTCACCCACCACCTCCTGTGATCTATCTCCCTG
Rhesus	GTCAGAGGTTGGAGGGGAGTGGTGAA--CGTTCATAAT---GAAAAGTCAACATGAAAG	CTTCTCGCGCATGGG---CGCTGGGCTCAGCCACCACCTCCTGTGCATCT---CCCTG	CTTCTCGCGCATGGG---CGCTGGGCTCAGCCACCACCTCCTGTGCATCT---CCCTG
Armadillo	GTCAG--GGTGAGGAGACAGTCTGAATCTTCTCTGT---GAGAA--AGTCAATATGAAAG	TCCCCTGGCAATGG---CTCTGGACCTTACCCACTACATCCTGTGTGCT---TTTTCA	TCCCCTGGCAATGG---CTCTGGACCTTACCCACTACATCCTGTGTGCT---TTTTCA
Cow	GCCAGAGGTAGAAAGAACCTTTGAATCAT--CAGT---AAACA--AGTTGGTGTGAAAG	CCTCTGGGCGACTCG---TCCCAGGCTTCACTACCCACATCCTGTGCATCT---CCCTG	CCTCTGGGCGACTCG---TCCCAGGCTTCACTACCCACATCCTGTGCATCT---CCCTG
Horse	GTCAGAGGTGGAGGGAGAGCTGTGAA--CATTCCTGT---AAAA--GATCGATGTGAAAG	CCCCATGGTGATGG---TGCTGGGCTCACCCACCACCTCCTGTGCATCT---TCCTG	CCCCATGGTGATGG---TGCTGGGCTCACCCACCACCTCCTGTGCATCT---TCCTG
Panda	GTCAGAGGTAGAGGAAGACT--GAA--CCTTCTCAT---AAAA--G-TCAGTGTGAAAG	CCCCATGGTGATGG---TGCTGGGCTCACCTACCACCTCCTGTGTGCT---CCTG	CCCCATGGTGATGG---TGCTGGGCTCACCTACCACCTCCTGTGTGCT---CCTG
Elephant	GTCAGAGGTGGAGGGAAAGCTGTGAA--CCTTTCTAT---AAGAA--AGTCAAGTGTGAAAG	TCCCATGGTGATGG---TGCTGGGCTCACCTACCACCTCCTGTGCATCT---CCTG	TCCCATGGTGATGG---TGCTGGGCTCACCTACCACCTCCTGTGCATCT---CCTG
Dog	GTCA--AAGTACAGGGAAGACTGTGAG--CCTTCTTAT---AAAA--GTCAGTGTGAAAG	CCCCATGGTGATGG---TGCTGGGCTCACCCACCACCTCCTGTGCATCT---CCCTG	CCCCATGGTGATGG---TGCTGGGCTCACCCACCACCTCCTGTGCATCT---CCCTG
Mouse	GTCAATGGTAAAGGGAGAGCTGTGAACTTTCTATTAATAAATAAATAAAT--GTGTGAAAG	TCCCATGGTGACTAG---TGTCAGGCTCACCCACCACCTCCTGTGCATCT---CCCTG	TCCCATGGTGACTAG---TGTCAGGCTCACCCACCACCTCCTGTGCATCT---CCCTG
Rat	ATCAATGGTAAAGGAGAGCTGTGAACTTTCTGTTTTAAAAAATAAATAAAT--GTGTGAAAG	TCCATGCTGATTAG---CGTTAGGCTCACCCACCACCTCCTGTGCATCT---CCCTG	TCCATGCTGATTAG---CGTTAGGCTCACCCACCACCTCCTGTGCATCT---CCCTG

Human	GCCCCCGGGCTC---AGTCCCCAC-----TGCTCAGCACTAGGCC--GGCAGAACTCTGA--	-GCGATG
Rhesus	GCCCCGTGGGCTC---AGTCCCCACCCGCTGCTCAGCACTCGACC--GGTAGAATCTGA--	-GCGATG
Armadillo	TCCTGCTCAGACACCGTGGTCTCCTCCAT-CTGCACAGT--TGGACCAGTGGAAAGGAGAT	-GCCATG
Cow	GCCCCGGGGCTCC---ATCTCCACCACAGCTGTCCA-CATTT-ACCAGGCAGAACTCTGA--	-GCGATG
Horse	GCCTACTGGCCTCA--GCTCTGCTCAGCTGCTCCACACTAGCCT--GGCAGAACTCTGT-	-GCTATG
Panda	GCCTCCTGGACTCC---GTCTCCACCAGCTGCTCCACATTCGTGC--GGTAGCATCAGT-	-GCAATG
Elephant	GCCTGCTGGACTTT--GCTCTCAACCAGCTGCTCCACATTCGCCT--GGCAGAACTCTGTG	TGCTATG
Dog	GCCTGCTGGGCTCA--GTCTCCACTCAGCTCCTCCATATGTGCTT--GGTATAATCAGT-	-GCTATG
Mouse	ACCCACTGGACTCA--ATCCACACACA-TAGTTCAGCACTGGTCTGGCCA--CTGG--	-TATG
Rat	ACCCACAGGATCA--GTCCACATACA-CTATTCAAGCAGCCGGACTGGCTA--CTGG--	-TATG

FIGURE S1

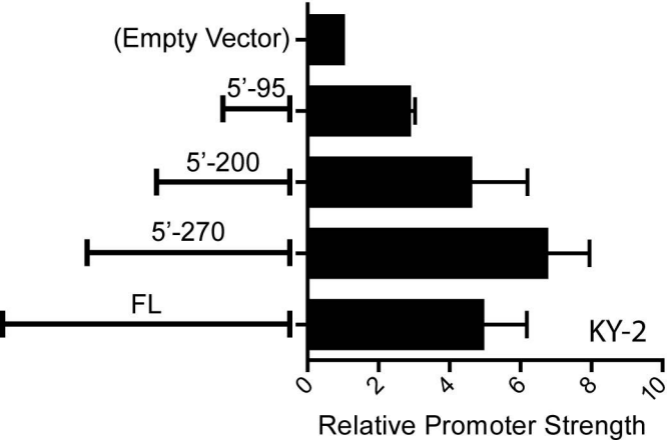
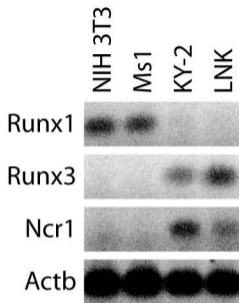
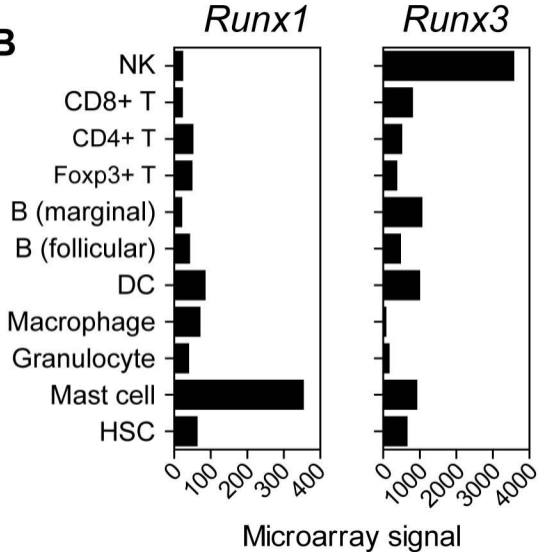


FIGURE S2

A**B****FIGURE S3**