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 Claustal2W. 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 μg vector, 0.1 μg pRL-TK, and 2 μg 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'aaggtacccgctggtgctcacccacc	5'aagctagctcgctcagattctgccgg
5'200-pGL4B	5'aaggtaccgtgtcagagggaccacgg	Same as above
5'270-pGL4B	5'aaggtaccgagaagttgacccagaaatgc	Same as above
5'395-pGL4B	5'aaggtacccctgttccagtatctcactg	Same as above
3'276-pGL4B	Same as above	5'aagctagcgactgctttcatcagaacg
3'196-pGL4B	Same as above	5'aagctagcatctgtggtctagccagc
3'105-pGL4B	Same as above	5'aagctagctacccagagctgactgtg
hPU.1-RT-PCR	5'ccaaacgcacgagtattacc	5'aagctctcgaactcgctgtg
hCJUN-RT-PCR	5'aaggaagctggagagaatcg	5'tgtttaagctgtgccacctg
hJUNB-RT-PCR	5'cgatctgcacaagatgaacc	5'tgctgaggttggtgtaaacg
hJUND-RT-PCR	5'gcctcatcatccagtccaac	5'tctgcttgtgtaaatcctccag
hCEBPa-RT-PCR	5'tggacaagaacagcaacgag	5'ttgtcactggtcagctccag
hMYB-RT-PCR	5'cagcccactgttaacaacgac	5'ctggctgagggacattgac
hMEIS1-RT-PCR	5'ggaggatcaaaatcagacagtg	5'tcctcctgaacgagtagatgc
hCP2-RT-PCR	5'ggtgaagagtatattccgtgtgg	5'attctgtctccaggtcggttc
hE2A-RT-PCR	5'gaagcagcagcacgtttg	5'gagaaggaggatgcagatgg
hRUNX1-qPCR	5'tcaggtttgtcggtcgaag	5'gcccatccactgtgattttg
hRUNX3-qPCR	5'gttcaacgaccttcgcttc	5'gtccacggtcaccttgatg
hRUNX3dist-RT-PCR	5'acctactcgccgaccttcat	Same as above
hRUNX3prox-RT-PCR	5'tattcccgtagacccaagca	Same as above
mNcr1-RT-PCR	5'tggctcttacaacgactatgc	5'agaagaagtagggtcggtaggtg
mRunx1-RT-PCR	5'tagcgagattcaacgacctc	5'atggtaggtggcaacttgtg
mRunx3-RT-PCR	5'ggttcaacgaccttcgattc	5'cggtggtaggtagccacttg
mActb-RT-PCR	5'aaggccaaccgtgaaaagat	5'gtggtacgaccagaggcatac
distRUNXmut	5'gagttgctggctagacgttagatgtgtcagaggg	5'ccctctgacacatctaacgtctagccagcaactc
proxRUNXmut	5'gctggtgctcacccacgttttcctgtgtatctatc	5'gatagatacacaggaaaacgtgggtgagcaccagc
hNCR1promoter-ChIP	5'ctgatgaaagcagtcaacgtg	5'gggccagggagatagatacac
h+8intergenic-ChIP	5'tatcttgacaagggctcacg	5'gcttcagcactgaatgatcc
hCD122promoter-ChIP	5'caagcaggtccctctaggtg	5'taaacggagtaaggggcttc
mNCR1promoter-ChIP	5'aaatgggttgcagactgagc	5'accattgacctaggacttcagg
mIfng-qPCR	5'cacggcacagtcattgaag	5'ccagttcctccagatatccaag
mPrf1-qPCR	5'gtgtcgcatgtacagttttcg	5'tgtggtaagcatgctctgtg
m18S-qPCR	5'gtaacccgttgaaccccatt	5'ccatccaatcggtagtagcg
mB2m-qPCR	5'catggctcgctcggtgacc	5'aatgtgaggcgggtggaactg
mCD43-qPCR	5'agtttctttgaccccttgg	5'ttctggaagcagtgctgatg
EMSA(enhancer)-WT	5'gttgctggctag**accaca**gatgtgtcagag	5'ctctgacacatc**tgtggt**ctagccagcaac
EMSA(enhancer)-MU	5'gttgctggctag**acgtta**gatgtgtcagag	5'ctctgacacatc**taacgt**ctagccagcaac
EMSA(essential)-WT	5'tggtgctcaccc**accact**tcctgtgtatct	5'agatacacagga**agtggt**gggtgagcacca
EMSA(essential)-MU	5'tggtgctcaccc**acgttt**tcctgtgtatct	5'agatacacagga**aaacgt**gggtgagcacca

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

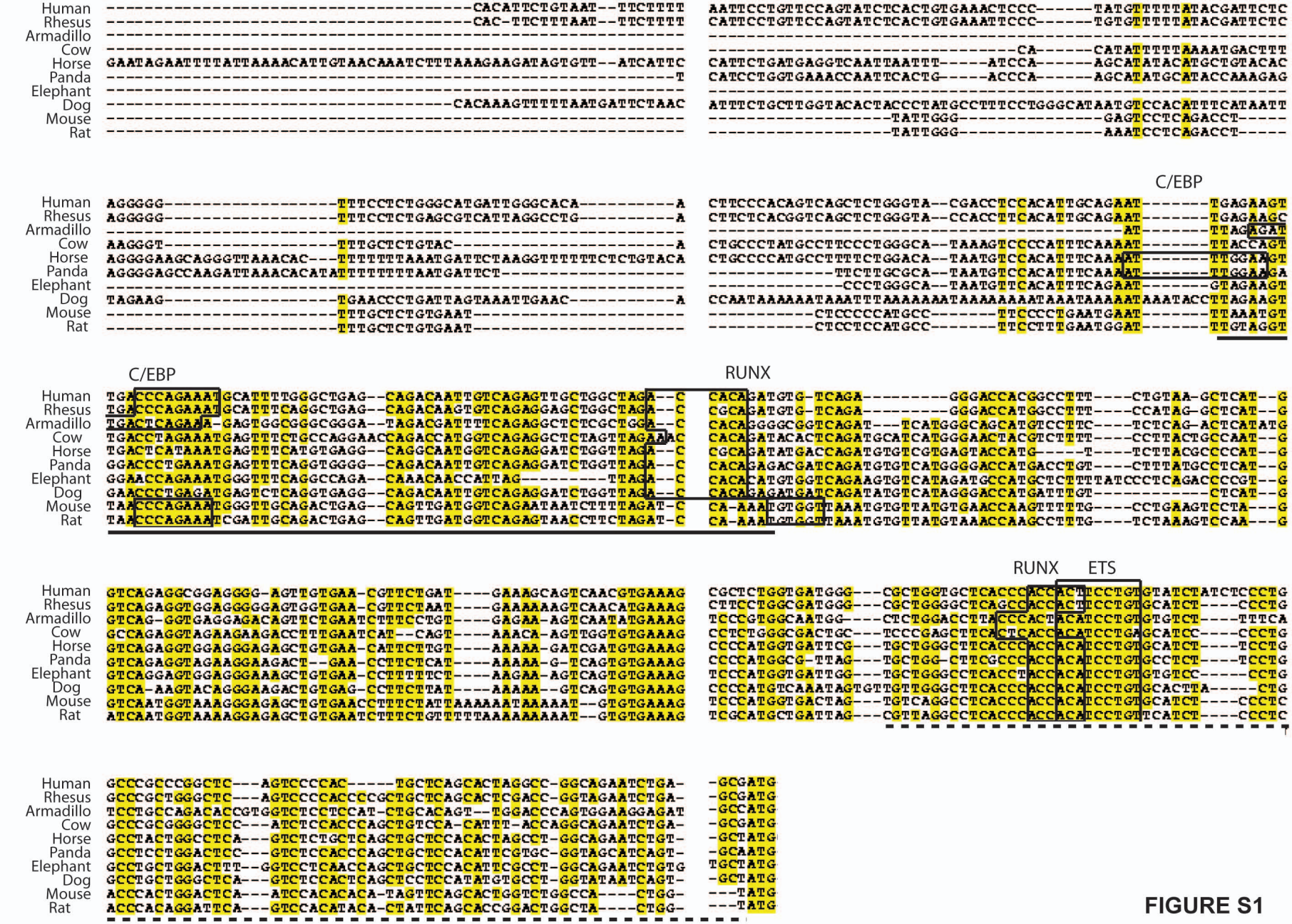
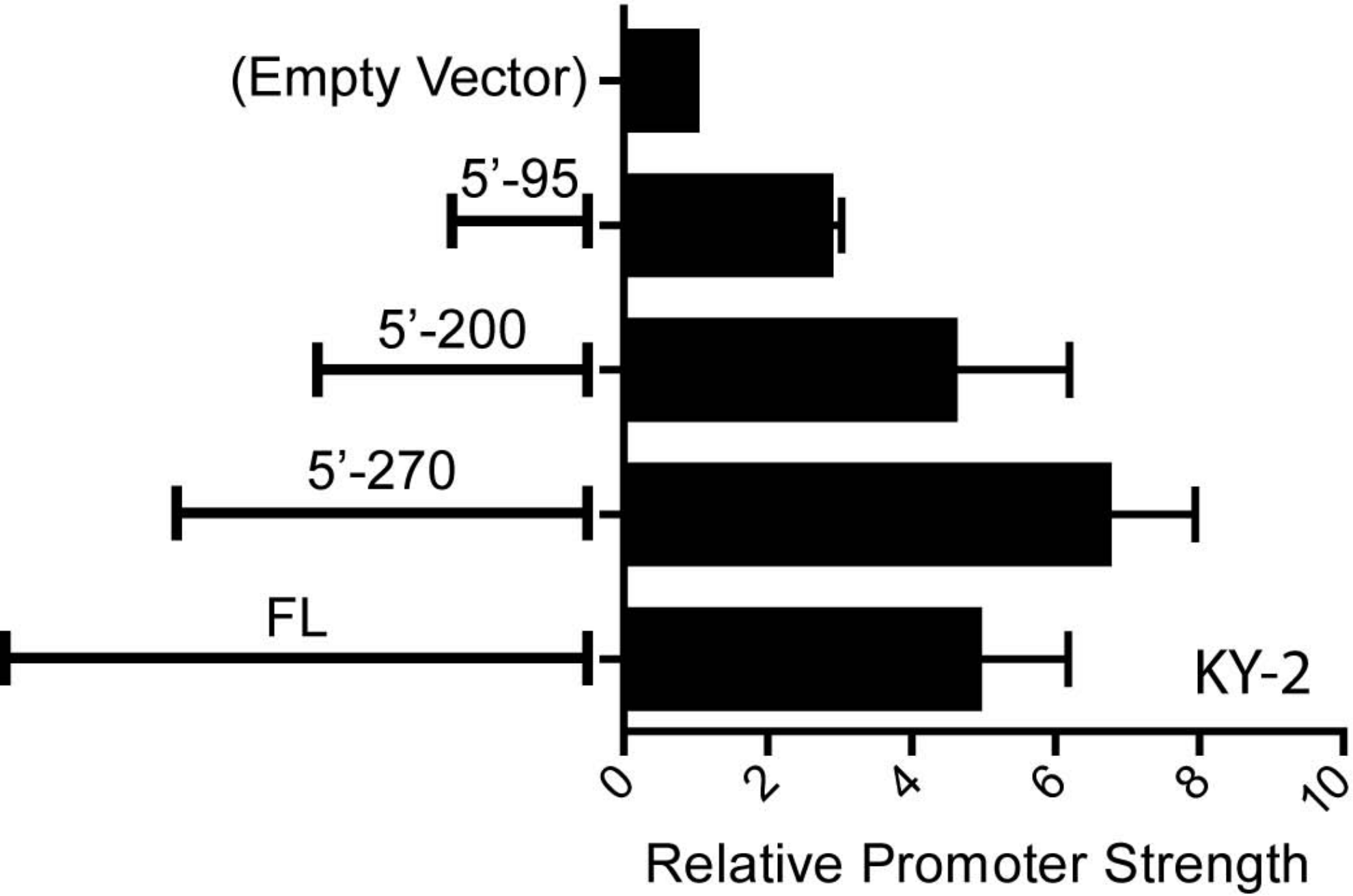

Human
Rhesus
Armadillo
Cow
Horse
Panda
Elephant
Dog
Mouse
Rat
C/EBP
C/EBP
RUNX
RUNX
ETS

FIGURE S1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5'-95
5'-200
5'-270
FL
KY-2
0
2
4
6
8
10
Relative Promoter Strength

FIGURE S2

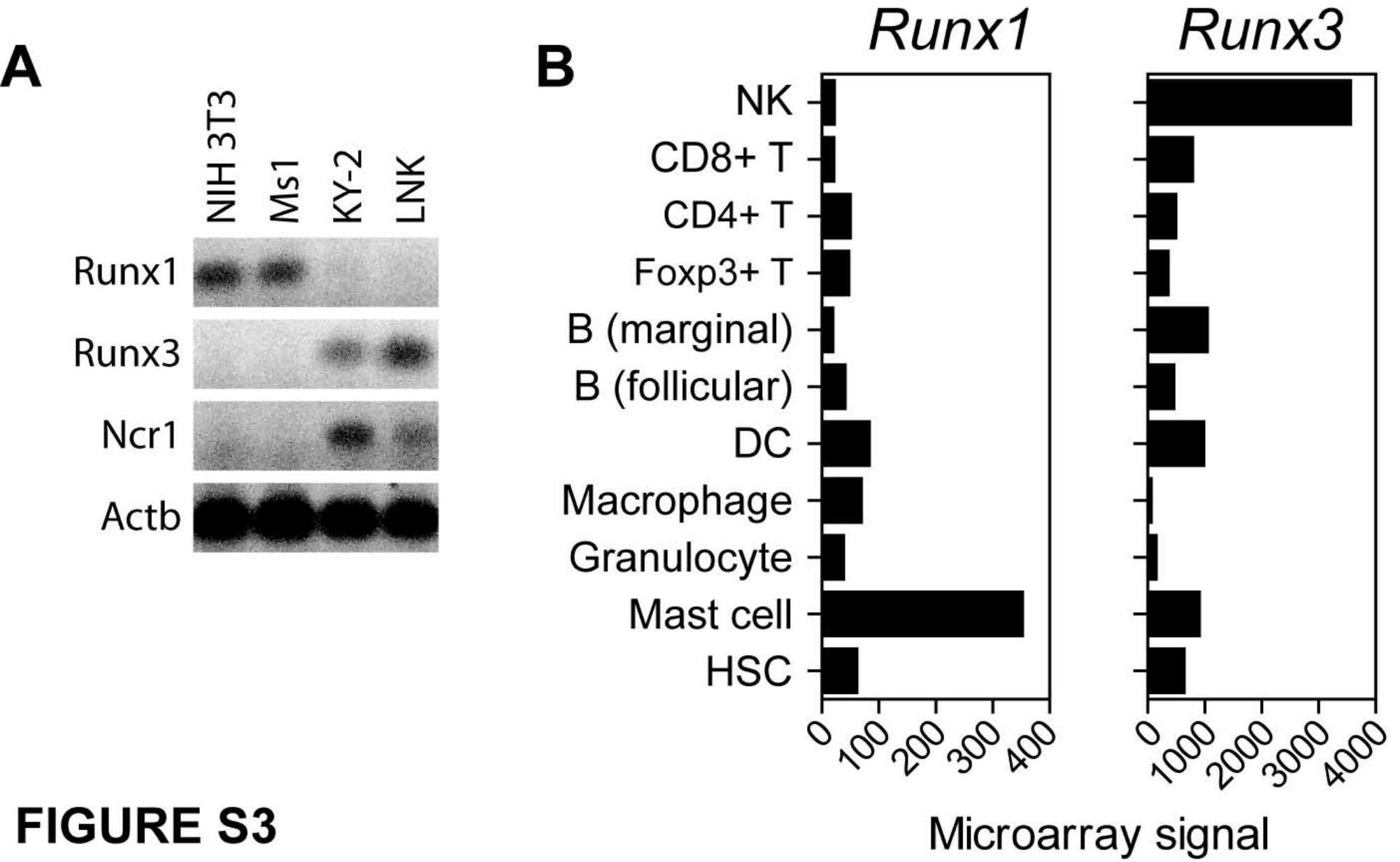
A
NIH 3T3
Ms1
KY-2
LNK
Runx1
Runx3
Ncr1
Actb
B
Runx1
Runx3
NK
CD8+ T
CD4+ T
Foxp3+ T
B (marginal)
B (follicular)
DC
Macrophage
Granulocyte
Mast cell
HSC
0
100
200
300
400
0
1000
2000
3000
4000
Microarray signal

FIGURE S3