Supplemental Material: Epistasis between TIFAB and miR-146a, neighboring genes in del(5q) MDS

Supplemental Methods

Mice and bone marrow transplantation.

Tifab$^{-/-}$ C57Bl/6 mice were described previously[1]. miR-146a$^{-/-}$ C57BL/6 mice were obtained from Dr. David Baltimore as previously described[2]. All animals were bred and housed in the Association for Assessment and Accreditation of Laboratory Animal Care-accredited animal facility of Cincinnati Children's Hospital Medical Center. For non-competitive BM transplantation, BM mononuclear cells (0.3-3x10^6) from C7BL/6 wild-type, *Tifab*$^{-/-}$, miR-146a$^{-/-}$, or *Tifab*$^{-/-}$;miR-146a$^{-/-}$ mice were injected *i.v.* into lethally-radiated (7.0 Gy and 4.75 Gy after 3 hours) syngeneic recipient Boy/J mice.

BM and PB analysis

For cytospins, 2.5-5x10^5 BM cells were spun onto slide at 300 rpm for 5 minutes and then stained with Wright-Giemsa. For BM sections, tibias were fixed in formalin and then stained with hematoxylin and eosin. Complete blood counts were performed on peripheral blood isolated from the tail vein every 4 weeks and analyzed using the Drew Scientific Hemavet 950.

Quantitative PCR analysis

RNA was extracted from cells and purified with Trizol Reagent (Life Technologies 15596-026) or Quick-RNA MiniPrep Kit (Zymo Research R1055). cDNA was generated using a high capacity RNA to cDNA kit (Life Technologies 4387406 or 4368814). qPCR was carried out using Taqman Master mix and probes (Life Technologies): *Tifab* (Mm0421026_m1) and *Gapdh* (Mm99999915_g1). qPCR was performed on an Applied Biosystems StepOne Plus Real-Time PCR System. miR-146a expression was performed as previously described[3].

Clonogenic progenitor assays

Hematopoietic clonogenic progenitor frequencies were determined by plating 2x10^5 BM cells/ml isolated from transplanted wild-type, *Tifab*$^{-/-}$, miR-146a$^{-/-}$, or *Tifab*$^{-/-}$;miR-146a$^{-/-}$ mice in methylcellulose media containing human erythropoietin, murine SCF, murine IL-3, and human IL-6 (Methocult M3434; Stem Cell Technologies). Colonies were scored after 10 days.

Flow cytometry analysis

Antibodies used for flow cytometric analysis of BM and PB cells are described previously[1]. All FACS analyses were performed on BD FACS Aria or Canto machines. FACS data analysis was performed using BD FACSDiva or FlowJo software.

Patient samples

Informed consent was obtained according to protocols approved by the review board of Cleveland Clinic. Diagnoses were reviewed at Cleveland Clinic and adapted, when required, to WHO 2008 criteria. For microarray and SNP analysis, BM mononuclear cells were isolated from MDS/AML patients, as previously described[2].

RNA sequencing analysis

Transcriptomes of lineage$^-$cKit$^+$ BM cells from wild-type (WT, n=3), *Tifab*$^{-/-}$ knockout (TIFAB-KO, n=3), *miR-146a*$^{-/-}$ (miR146a-KO, n=3), *Tifab*$^{-/-;}$ *miR-146a*$^{-/-}$ mice (dKO, n=4) were compared by standard RNA-seq analysis methods. Total RNA was amplified using the Ovation RNA-Seq System v2 (NuGEN) according to the manufacturer's protocol. The libraries were prepared with the Nextera XT DNA Sample Preparation kit (Illumina Technologies). 1 ng of cDNA was suspended in Tagment DNA Buffer, and tagmentation (fragmentation and tagging with the adaptors) was performed with the Nextera enzyme (Amplicon Tagment Mix), incubating at 55°C for 10 min. NT Buffer was then added to neutralize the samples. Libraries were prepared by PCR with the Nextera PCR Master Mix, and 2 Nextera Indexes (N7XX, and N5XX) according to the following program: one cycle of 72°C for 3min, one cycle of 98°C for 30s, 12 cycles of 95°C for 10s, 55°C for 30s, and 72°C for 1min, and one cycle of 72°C for 5min. The purified cDNA was captured on an Illumina flow cell for cluster generation. Libraries were sequenced on the Illumina HiSeq2500 following the manufacturer's protocol. Paired-end reads were aligned to UCSC mm10 genome (downloaded from Illumina's iGenomes repository; https://support.illumina.com/sequencing/sequencing_software /igenome.html) using TopHat[4] and mouse mm10-refseq gene GTF file. FeatureCounts was used for read counting[5]. Differentially expressed genes were predicted by three independent methods including limma/voom[6,7], edgeR[8], and DESeq2[9]. FDR < 0.05 and fold change > 2x were used for default cutoffs.

Synergy gene analysis

To examine synergistic effects, we adopted a concept used in McMurray *et al*[10]. Briefly, *a* = fold change value for a given gene in the miR-146a$^{-/-}$ group, *b* = fold change value for the same gene in the Tifab$^{-/-}$ group, and *d* = mean expression value for this gene in the dKO group. Then synergy score was calculated using $(a+b)/d$ if $d>1$ and $d/a + d/b$ if $d<1$.

Supplemental References

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4. Trapnell, C., Pachter, L. & Salzberg, S.L. TopHat: discovering splice junctions with RNA-Seq. *Bioinformatics* **25**, 1105-11 (2009).
5. Liao, Y., Smyth, G.K. & Shi, W. featureCounts: an efficient general purpose program for assigning sequence reads to genomic features. *Bioinformatics* **30**, 923-30 (2014).
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7. Ritchie, M.E. *et al.* limma powers differential expression analyses for RNA-sequencing and microarray studies. *Nucleic Acids Res* **43**, e47 (2015).
8. Robinson, M.D., McCarthy, D.J. & Smyth, G.K. edgeR: a Bioconductor package for differential expression analysis of digital gene expression data. *Bioinformatics* **26**, 139-40 (2010).
9. Love, M.I., Huber, W. & Anders, S. Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. *Genome Biol* **15**, 550 (2014).
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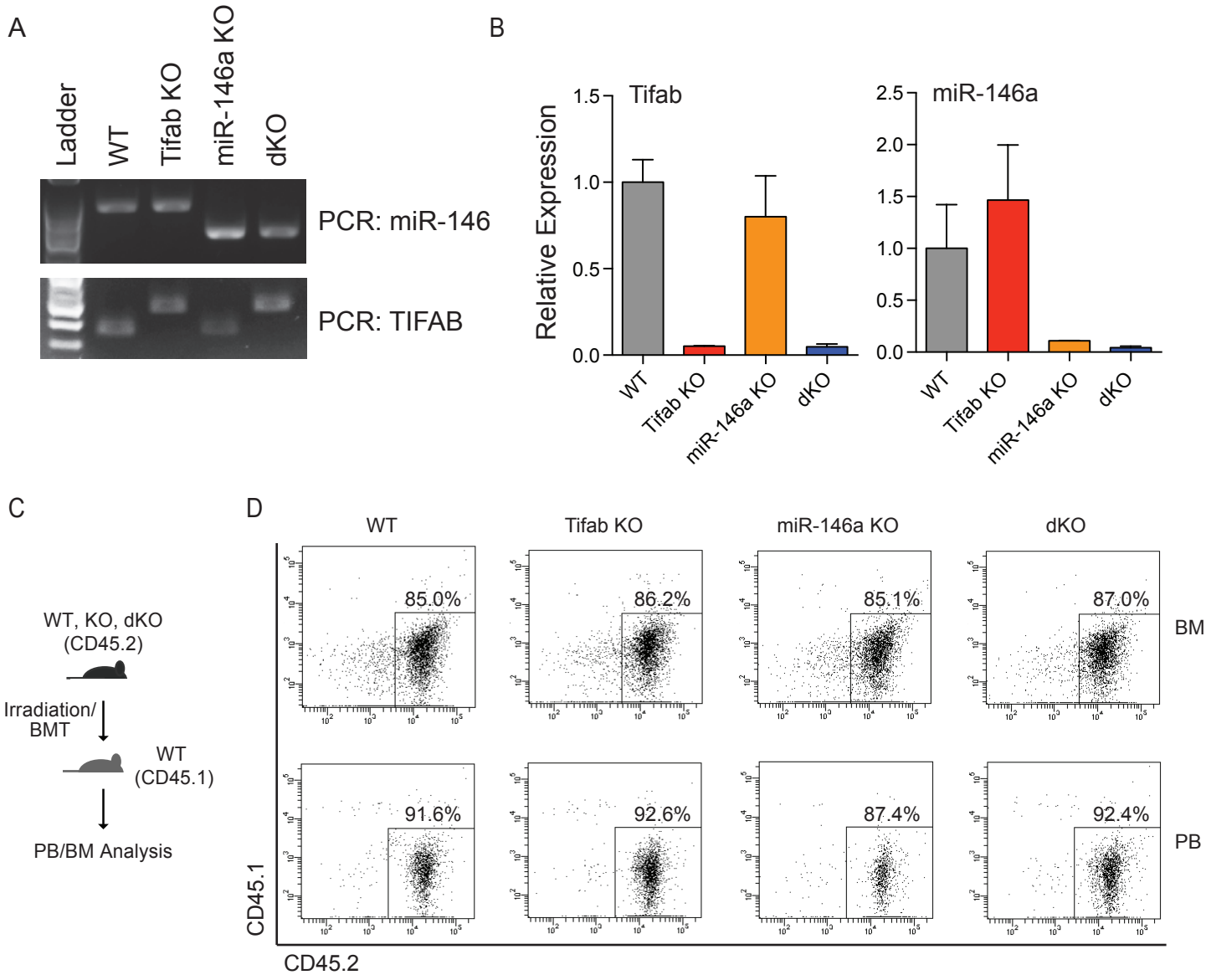
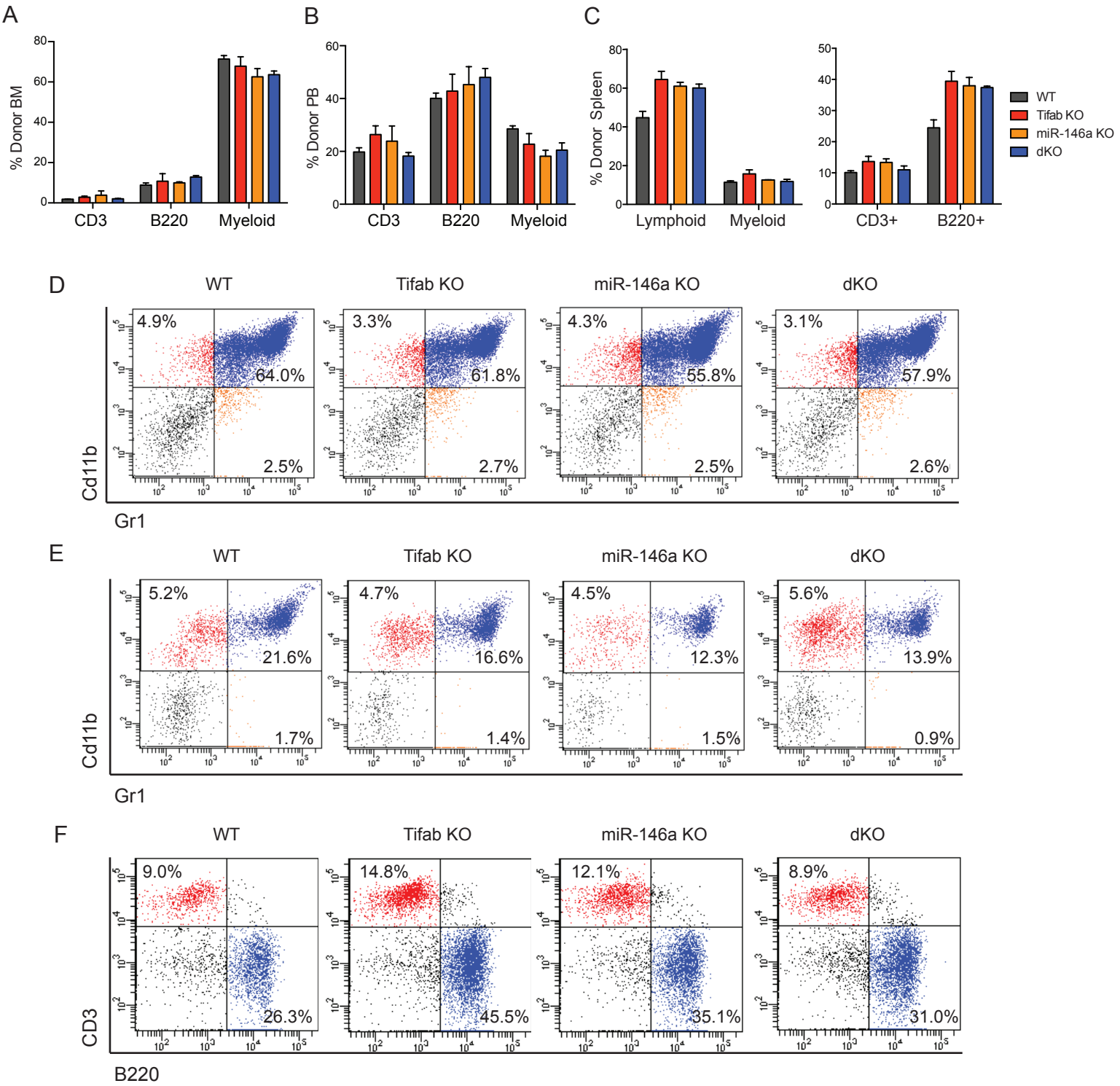
Supplemental Figure 1



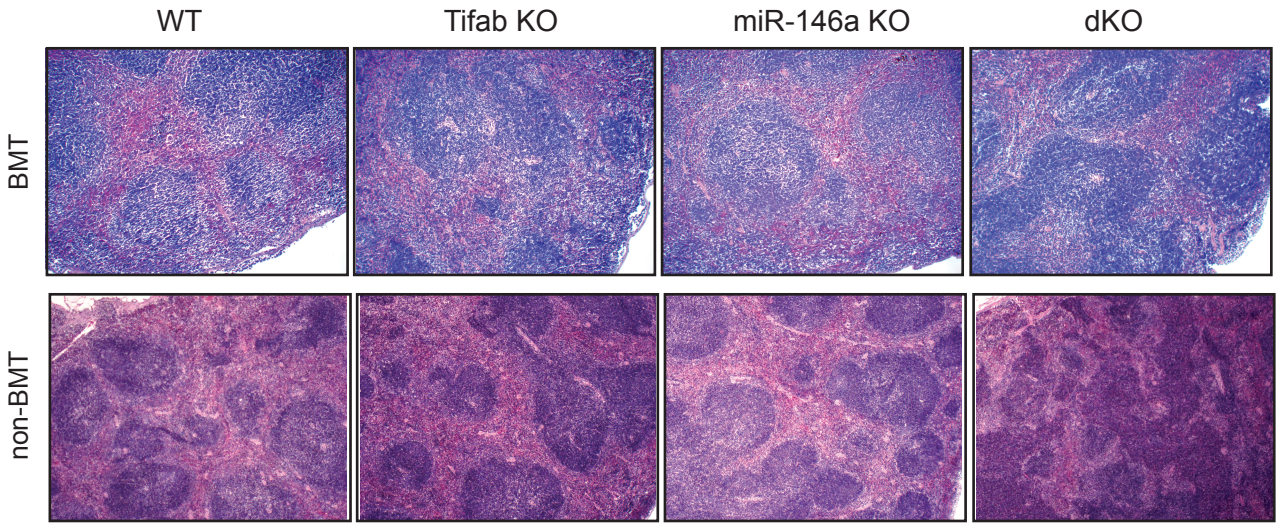
Generation of Tifab and miR-146a double knockout mice. (A) PCR was performed using primers to detect wild-type or deleted miR-146a and Tifab alleles on genomic DNA. (B) Quantitative RT-PCR was performed to measure Tifab mRNA or miR-146a from BM mononuclear cells. (C) Overview of transplantation of BM cells from WT, Tifab KO, miR-146a KO, and dKO (CD45.2) mice into lethally-irradiated WT CD45.1 mice. (D) Proportion of donor cells in the peripheral blood and BM of recipient mice (n = 3 mice evaluated per group).

Supplemental Figure 2



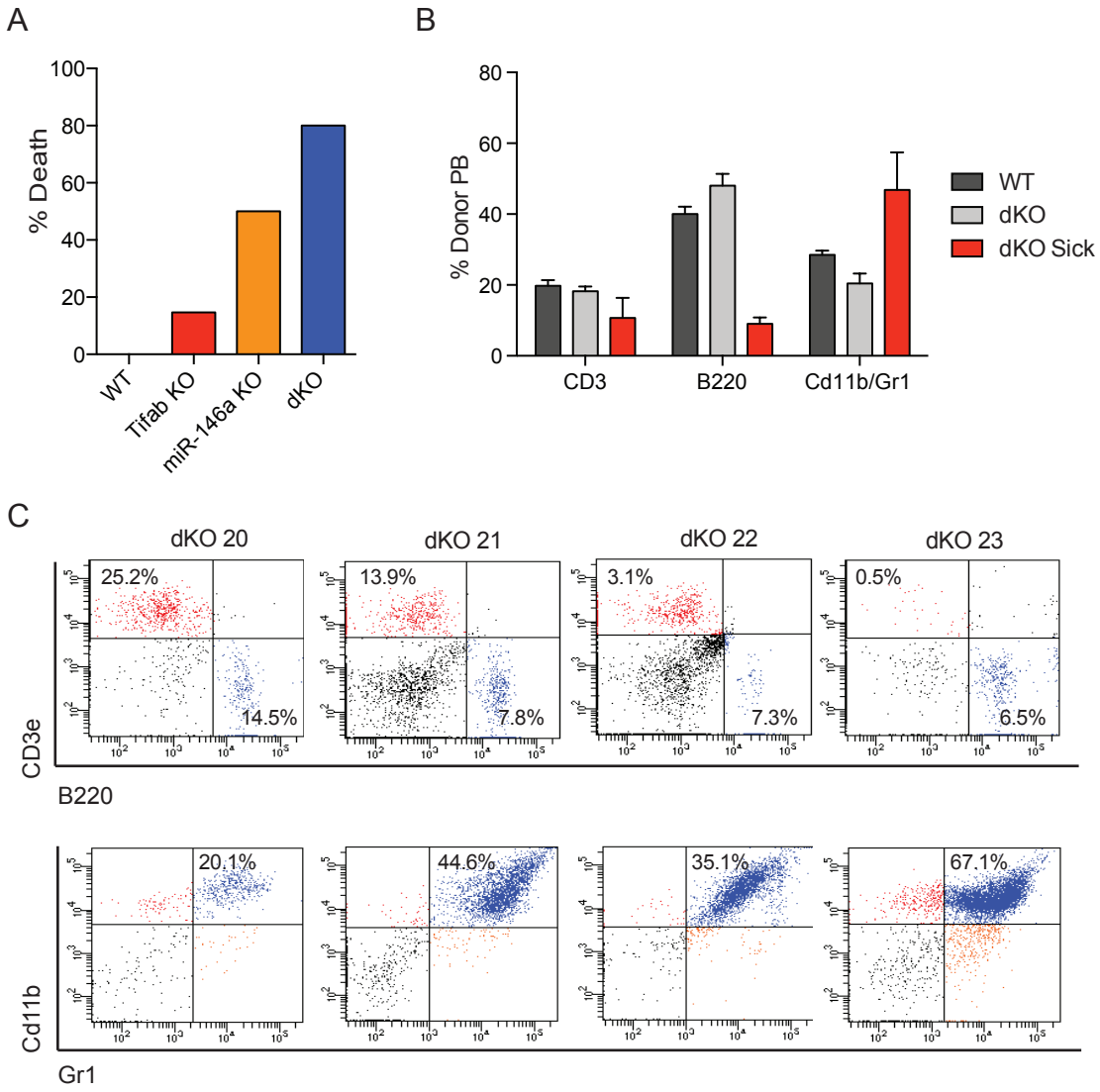
Immunophenotypic analysis of bone marrow, peripheral blood, and spleen. (A-C) Flow cytometric analysis of myeloid (CD11b, Gr1) and lymphoid (CD3, B220) proportions within the BM (A), PB (B), and spleen (C) of transplanted mice. (D-F) Representative flow cytometric plots from BM (D), PB (E), and spleen (F). Percent values within dot blots are calculated from the parent gate.

Supplemental Figure 3



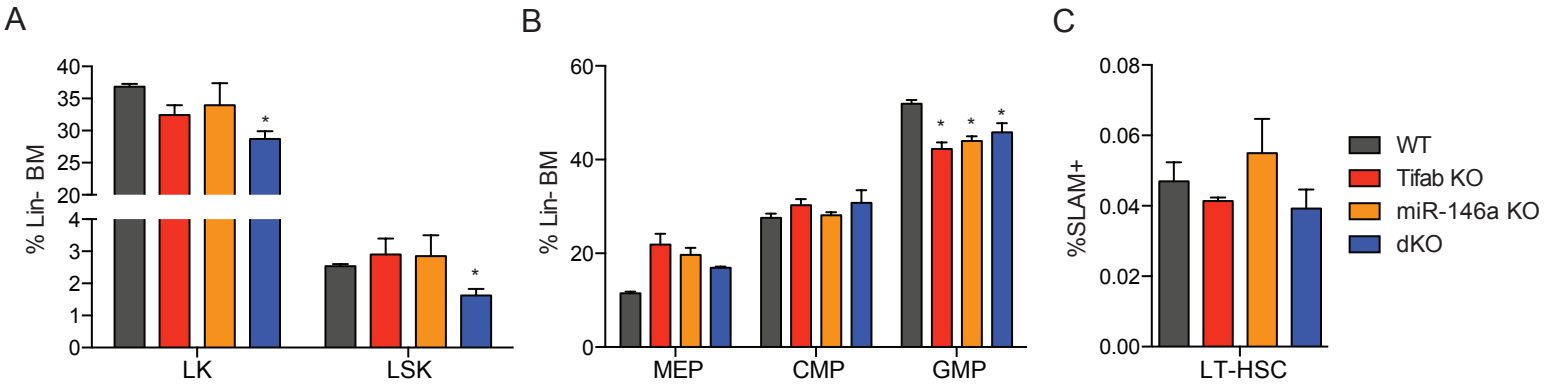
Morphological analysis of spleens. H&E-stained spleen from representative mice prior (non-BMT) or after transplantation (BMT).

Supplemental Figure 4.



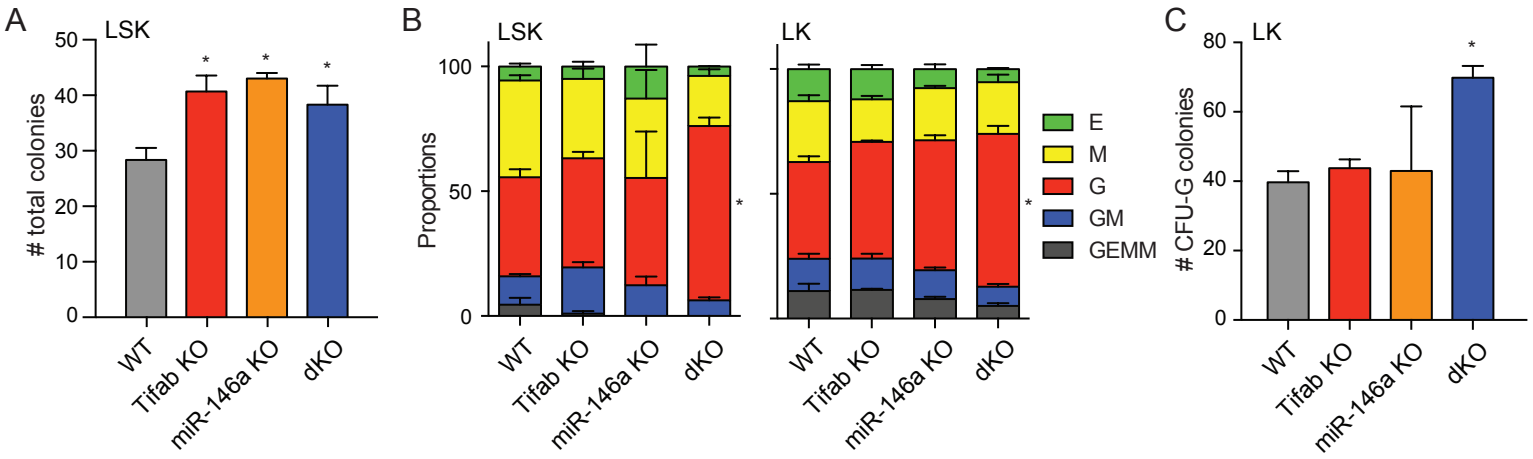
Analysis of moribund mice. (A) Percent of mice became moribund after transplantation. (B) Immunophenotype analysis of PB from WT, dKO, and moribund dKO mice. (C) Representative flow cytometric blots of PB cells from four moribund dKO mice. Percent values within dot blots are calculated from the parent gate.

Supplemental Figure 5.



Analysis of hematopoietic stem/progenitor proportions in the BM. (A) LK and LSK proportions in BM from mice transplanted with WT, Tifab-/-, miR-146a-/-, or dKO BM cells (n = 3). (B) MEP, CMP, and GMP proportions in BM from mice transplanted with WT, Tifab-/-, miR-146a-/-, or dKO BM cells (n = 3). (C) LT-HSC (LSK+ CD150+CD48-) proportions in BM from mice transplanted with WT, Tifab-/-, miR-146a-/-, or dKO BM cells (n = 3). *, $P < 0.05$; Student's t-test. In all cases, statistical comparisons were made to WT mice.

Supplemental Figure 6.



Analysis of hematopoietic stem/progenitor colony-forming function. (A) Numbers of colony-forming cells in methylcellulose from LSK cells from WT, Tifab-/-, miR-146a-/-, or dKO transplanted mice (n = 3). (B) Proportion of colony-forming cells in methylcellulose from LSK or LK isolated from WT, Tifab-/-, miR-146a-/-, or dKO transplanted mice (n = 3). GM, granulocyte-monocyte; G, granulocyte; M, monocyte; E, erythroid (BFU-E). (C) Numbers of CFU-G from LK cells from WT, Tifab-/-, miR-146a-/-, or dKO transplanted mice (n = 3).*, $P < 0.05$; Student's t-test as compared to WT.

Supplemental Table 1. Differentially expressed genes in LK.

Tifab KO vs WT

Gene	Fold Change (Log2)	P value	FDR
Tifab	-1.94	6.34E-15	1.53E-10
Prom1	2.27	1.58E-10	1.90E-06
Dio2	1.13	1.81E-07	0.001
Dntt	-1.37	1.96E-07	0.001
C3	1.15	2.36E-07	0.001
Slc15a2	1.83	7.26E-07	0.003
H2afy	-1.05	9.67E-07	0.003
Mtus1	1.40	9.95E-07	0.003
Pglyrp1	1.27	1.33E-06	0.004
Nucb2	1.22	1.69E-06	0.004
Prkcb	1.14	1.76E-06	0.004
Ms4a3	1.22	4.32E-06	0.009
Syce1	-8.11	5.16E-06	0.010
F630028O10Rik	1.12	7.48E-06	0.013
Ncam1	1.49	8.51E-06	0.014
B4galt6	1.50	1.17E-05	0.018
Fcer1a	1.17	1.44E-05	0.020
Gfi1	1.24	2.20E-05	0.028
Mrc1	1.90	2.26E-05	0.028
Axl	1.76	2.32E-05	0.028
Vcam1	1.86	3.11E-05	0.034
Gem	-1.19	3.21E-05	0.034
Mras	2.29	3.29E-05	0.034
Prtn3	1.07	3.48E-05	0.035
Col5a1	-1.07	3.84E-05	0.037
Tex11	5.92	4.04E-05	0.037
Epx	1.31	4.24E-05	0.038
Ear1	1.44	4.61E-05	0.040
Gzmb	1.00	5.55E-05	0.045
Igf1	2.62	5.58E-05	0.045
Enah	1.63	5.94E-05	0.046
Gda	1.06	6.19E-05	0.047
Slmo1	-3.11	6.38E-05	0.047
Lrguk	-6.06	6.83E-05	0.048
Igsf6	0.82	7.05E-05	0.049
C1qb	2.80	7.37E-05	0.049
Abcc2	1.85	7.49E-05	0.049
B4galt4	0.90	7.70E-05	0.049

miR-146a KO vs WT

Gene	Fold Change (Log2)	P value	FDR
Lyrm7	3.10	3.19E-32	7.69E-28
Cmss1	-2.46	9.72E-22	1.17E-17
Gzmb	1.66	5.90E-17	4.73E-13
Olfr56	-8.25	1.65E-16	9.94E-13
Gm2a	-1.30	1.71E-15	8.21E-12
Dio2	1.34	2.79E-15	1.12E-11
Wdfy1	1.51	4.76E-14	1.51E-10
Vps13c	1.12	5.34E-14	1.51E-10
F630028O10Rik	1.26	5.64E-14	1.51E-10
1700112E06Rik	-2.66	9.81E-13	2.36E-09
Prkcb	1.43	1.75E-12	3.84E-09
Pik3r5	1.42	1.17E-11	2.36E-08
Cyp11a1	2.64	5.34E-11	9.88E-08
Mtus1	1.45	1.34E-10	2.31E-07
Irgm2	1.86	2.09E-10	3.35E-07
Nr4a1	-1.54	8.01E-10	1.20E-06
Efcab7	1.37	1.30E-09	1.84E-06
Egr3	-1.62	8.00E-09	1.07E-05
Erdr1	1.11	1.55E-08	1.96E-05
Iigp1	2.52	2.87E-08	3.46E-05
Dntt	-1.23	3.62E-08	4.15E-05
Serpina3g	0.98	5.46E-08	5.97E-05
Zbtb16	1.05	8.85E-08	9.26E-05
Nlrc5	0.94	1.01E-07	0.0001
Tln2	2.15	1.12E-07	0.0001
Gm12250	1.51	1.60E-07	0.0001
Skp2	1.02	1.73E-07	0.0002
Smim3	-1.04	1.76E-07	0.0002
C3	0.99	2.45E-07	0.0002
Atrn	0.75	3.00E-07	0.0002
Tsc22d3	-1.25	3.07E-07	0.0002
Irgm1	1.03	3.74E-07	0.0003
Fcer1a	1.02	4.11E-07	0.0003
Rplp1	-0.83	6.05E-07	0.0004
2810417H13Rik	-0.77	6.45E-07	0.0004
Nrg1	2.57	9.18E-07	0.0006
Clec5a	1.37	9.46E-07	0.0006
Dock9	0.99	1.05E-06	0.0007
Stk17b	-0.82	1.14E-06	0.0007
Ptpb	-1.21	1.30E-06	0.0008
Cleo4a3	4.99	1.33E-06	0.0008
Mir3064	1.70	1.79E-06	0.0010
Lpgat1	0.80	1.87E-06	0.0010
Tgtp1	2.58	1.93E-06	0.0011
Hist1h2bc	-1.00	2.02E-06	0.0011
Hmgn5	-0.80	2.07E-06	0.0011
Camk2b	-3.82	2.21E-06	0.0011
Rab6b	-1.33	2.41E-06	0.0012
Slc31a2	1.08	2.55E-06	0.0013
B4galt6	1.31	2.99E-06	0.0014
Gstm5	-0.84	3.01E-06	0.0014
H1f0	-0.81	3.15E-06	0.0015
Gm1966	0.69	3.45E-06	0.0016
Syce1	-8.00	3.87E-06	0.0017
Atp11a	0.81	4.10E-06	0.0018
App	0.86	4.18E-06	0.0018
Sdf2l1	0.82	4.75E-06	0.0020
Pf4	-0.73	4.84E-06	0.0020
Tmem9	-1.00	5.00E-06	0.0020
Znf41-ps	1.02	6.03E-06	0.0024
Sowahc	1.02	6.05E-06	0.0024
Rpl19	-0.81	7.26E-06	0.0028
Igtp	0.91	7.50E-06	0.0029
Mras	2.15	7.88E-06	0.0030
Ttll4	0.97	8.04E-06	0.0030
Fndc3a	0.68	8.20E-06	0.0030
Ikzf3	1.67	9.95E-06	0.0036
Gpr84	2.50	1.03E-05	0.0036
Rps4x	-0.92	1.12E-05	0.0039
Aias1	0.74	1.14E-05	0.0039
R3hdm4	-0.69	1.19E-05	0.0040
H2-Q5	0.85	1.24E-05	0.0042
Bpifc	-0.97	1.27E-05	0.0042
Ccdc69	-1.05	1.28E-05	0.0042
Rpl23	-0.67	1.32E-05	0.0042
Gpr35	2.68	1.33E-05	0.0042
Tmem50b	0.77	1.45E-05	0.0045
R3hcc1	-1.25	1.51E-05	0.0046
Hist1h3d	1.42	1.59E-05	0.0048
Slamf7	1.40	1.74E-05	0.0052
Senp5	0.80	1.80E-05	0.0053
H3f3b	-0.72	1.83E-05	0.0053
Rpl14	-0.94	1.87E-05	0.0053
Pde2a	0.98	1.89E-05	0.0053
Adpgk	0.78	1.91E-05	0.0053
Tnfaip3	-0.85	1.91E-05	0.0053
Lyst	0.63	2.04E-05	0.0056
Ahnak	-0.81	2.14E-05	0.0058
Ear1	1.65	2.14E-05	0.0058
Parp14	0.74	2.23E-05	0.0060
Slfn9	0.82	2.26E-05	0.0060
Ebf1	2.06	2.39E-05	0.0062
Rps14	-0.70	2.40E-05	0.0062
Klhl42	1.11	2.46E-05	0.0063
Dusp8	-2.31	2.71E-05	0.0068
Eml5	0.82	2.72E-05	0.0068
Ccnt2	0.65	2.88E-05	0.0072
Iglf1	2.13	2.96E-05	0.0073
Tmsb10	-0.76	3.08E-05	0.0075
Per1	-1.03	3.21E-05	0.0077
Olfr1396	-6.14	3.63E-05	0.0087
Srsf5	-0.73	3.72E-05	0.0088
Gpr128	0.67	3.77E-05	0.0088
Pou2af1	1.77	3.97E-05	0.0092
Neo1	3.34	4.11E-05	0.0093
Kcna2	4.11	4.12E-05	0.0093
Acvr1b	0.97	4.15E-05	0.0093
Wdr83os	-0.70	4.38E-05	0.0097
Fam46a	0.61	4.39E-05	0.0097
4930503L19Rik	-0.82	4.68E-05	0.0102
Gm12185	2.03	4.72E-05	0.0102
Abca13	1.21	4.74E-05	0.0102
Tnfsf15	3.46	5.03E-05	0.0107
Stat1	0.63	6.11E-05	0.0127
G2e3	0.76	6.12E-05	0.0127
Fosb	-0.99	6.13E-05	0.0127
Cp	1.72	6.22E-05	0.0128
Igsf6	0.75	6.52E-05	0.0132
Tifab	0.66	6.54E-05	0.0132
Cdkn1a	-0.89	7.31E-05	0.0147
Dmxl2	0.74	7.41E-05	0.0147
Rps10	-0.80	7.90E-05	0.0156
Csrnp1	-0.94	7.99E-05	0.0156
Cers6	0.61	8.06E-05	0.0156
Zfp518b	0.87	8.20E-05	0.0158
Fbx	0.87	8.36E-05	0.0160
Otud4	0.59	8.49E-05	0.0160
Rasgef1b	-0.99	8.53E-05	0.0160
Atp8b4	0.68	8.76E-05	0.0163
1110038B12Rik	0.65	8.86E-05	0.0163
Crip1	-0.67	8.91E-05	0.0163
Shprh	0.65	8.93E-05	0.0163
Ripk1	0.60	9.10E-05	0.0164
Tnfrsf12a	-1.35	9.13E-05	0.0164
S1pr3	0.93	9.35E-05	0.0167
Rrm2	0.58	9.50E-05	0.0168
Bhlhe40	-1.28	9.75E-05	0.0171
Rpl32	-0.74	0.0001003	0.0174

dKO vs WT

Gene	Fold Change (Log2)	P value	FDR
Lyrm7	3.15	1.67E-28	4.03E-24
Wdfy1	2.23	2.89E-26	3.48E-22
Tifab	-2.79	2.36E-18	1.89E-14
Gm2a	-1.34	1.17E-15	7.01E-12
Fosb	-1.31	5.48E-14	2.64E-10
Tsc22d3	-1.65	7.32E-13	2.94E-09
Olfr56	-5.38	2.38E-12	8.19E-09
H2afy	-1.03	1.73E-10	5.19E-07
Rplp1	-1.16	3.82E-09	1.02E-05
Emp1	-2.06	1.93E-08	4.66E-05
Vps13c	0.90	4.14E-08	9.05E-05
G2e3	1.11	5.42E-08	0.0001
Crip1	-1.19	8.04E-08	0.0001
Ahnak	-0.90	1.71E-07	0.0003
Lyplal1	3.23	2.28E-07	0.0004
Gdpgp1	1.61	3.11E-07	0.0005
Ncam1	1.82	3.47E-07	0.0005
Skp2	0.93	5.06E-07	0.0007
4930433N12Rik	5.09	6.69E-07	0.0008
F630028O10Rik	1.03	6.74E-07	0.0008
Smim3	-1.02	9.77E-07	0.0011
Klf10	-0.93	1.11E-06	0.0012
Cdkn1a	-1.24	1.22E-06	0.0013
Klhl42	1.13	1.91E-06	0.0019
Rplp2	-0.82	1.95E-06	0.0019
G530011O06Rik	1.11	2.33E-06	0.0022
Perp	-1.77	3.22E-06	0.0029
Dio2	1.08	5.11E-06	0.0043
Zfp354a	4.73	5.29E-06	0.0043
Bhlhe40	-1.58	5.33E-06	0.0043
Slc26a2	1.07	6.51E-06	0.0051
Rps15	-0.67	6.94E-06	0.0052
Klf4	-0.91	7.10E-06	0.0052
Ubc	-1.02	8.09E-06	0.0057
Kcnj2	1.26	9.09E-06	0.0062
Rpl37a	-0.78	9.39E-06	0.0062
Erdr1	1.05	9.47E-06	0.0062
Vcam1	2.11	1.18E-05	0.0074
Cleo4a3	6.07	1.48E-05	0.0089
Csrnp1	-0.90	1.49E-05	0.0089
Rps26	-0.73	1.56E-05	0.0092
Slc31a2	1.01	1.65E-05	0.0094
Per1	-1.22	1.78E-05	0.0100
Rps14	-0.92	1.97E-05	0.0108
Vamp8	-0.93	2.02E-05	0.0108
Olfr1396	-6.16	2.19E-05	0.0114
Tmem9	-0.95	2.32E-05	0.0119
R3hdm4	-0.77	2.39E-05	0.0120
Mtus1	1.29	2.65E-05	0.0129
Pisd-ps3	0.96	2.67E-05	0.0129
Ccdc107	-1.46	2.74E-05	0.0129
Klf6	-0.77	2.97E-05	0.0134
Adpgk	0.85	3.06E-05	0.0134
Rpl23	-0.78	3.07E-05	0.0134
Rc3h2	0.76	3.14E-05	0.0134
Irgm1	1.13	3.14E-05	0.0134
Rasa2	0.99	3.18E-05	0.0134
Kcna2	4.22	3.28E-05	0.0135
Aoah	1.13	3.32E-05	0.0135
Stard6	1.50	3.42E-05	0.0137
Tgtp1	3.70	3.56E-05	0.0141
Itga1	1.20	3.72E-05	0.0143
Nrg1	3.13	3.73E-05	0.0143
Clec5a	1.69	4.05E-05	0.0152
Dusp16	-0.81	4.11E-05	0.0152
Tln2	1.83	4.17E-05	0.0152
Cyp11a1	2.60	4.23E-05	0.0152
Eml5	0.90	4.29E-05	0.0152
Abca13	2.23	4.68E-05	0.0163
Trib1	-0.68	5.30E-05	0.0182
Pik3c2a	0.82	5.49E-05	0.0186
Mid1	0.85	5.64E-05	0.0189
Pcdha8	5.70	5.73E-05	0.0189
Cmss1	-0.99	6.01E-05	0.0196
Hist1h3d	1.79	6.49E-05	0.0208
Rpl36	-0.82	6.74E-05	0.0213
Atrn	0.79	7.17E-05	0.0224
Gpr128	0.78	7.60E-05	0.0232
Bpifc	-1.01	7.62E-05	0.0232
Slfn9	0.83	7.87E-05	0.0237
Zbtb16	0.90	8.17E-05	0.0238
Oit3	-2.81	8.19E-05	0.0238
Rpl24	-0.85	8.21E-05	0.0238
Zdhhc21	0.65	8.55E-05	0.0245
Ctla2a	-0.62	8.91E-05	0.0252
Ier2	-0.81	8.99E-05	0.0252
H3f3b	-0.56	0.0001161	0.0321
Mat2a	0.60	0.0001187	0.0325
Rassf1	-0.80	0.0001226	0.0332
Prkcb	1.56	0.0001296	0.0347
Fam195b	-1.07	0.0001318	0.0347
Rasgef1b	-0.83	0.0001326	0.0347
Neto2	0.95	0.0001345	0.0348
Ier5	-1.07	0.0001531	0.0392
Pink1	-0.91	0.0001589	0.0403
Wdr76	0.82	0.0001699	0.0426
Slk1	-0.66	0.0001773	0.0433
Zkscan8	0.72	0.0001777	0.0433
Errfi1	-1.42	0.0001783	0.0433
Fcgr4	3.68	0.0001846	0.0444
Nhp2	-0.75	0.0001927	0.0458
Atp5e	-0.73	0.0001956	0.0458
Kcnj5	-0.96	0.0001959	0.0458
Ltf	4.08	0.0002005	0.0464
Serpinb8	-2.40	0.000203	0.0465
Ifitm2	-0.99	0.000207	0.0470
Rps25	-0.68	0.0002134	0.0480
Pf4	-0.84	0.00022	0.0490
Stk17b	-0.63	0.0002251	0.0497

Epx	1.20	0.0001003	0.0174
Fndc7	3.31	0.0001023	0.0176
Rps3a1	-0.61	0.0001044	0.0177
Vwf	-0.68	0.0001045	0.0177
Afap1	0.86	0.0001057	0.0178
Car12	0.90	0.0001065	0.0178
Car1	-0.62	0.0001076	0.0178
Gpatch4	-0.86	0.0001077	0.0178
Gbp7	0.60	0.000111	0.0181
Gm1821	-1.39	0.0001116	0.0181
Ndrg1	-0.59	0.0001178	0.0189
Ier2	-0.75	0.0001178	0.0189
Gdpgp1	1.08	0.0001185	0.0189
Commd4	-0.88	0.0001196	0.0189
Zfp346	0.76	0.0001259	0.0197
Homez	0.93	0.0001259	0.0197
Rpl37a	-0.62	0.0001281	0.0199
Calr	0.67	0.0001292	0.0199
Gda	0.79	0.0001356	0.0208
Lmna	-1.04	0.0001408	0.0214
Rpl13	-0.62	0.0001451	0.0220
Vamp8	-0.67	0.0001468	0.0220
Fancm	0.65	0.0001478	0.0220
Tlr3	1.60	0.0001486	0.0220
Neurl3	-0.74	0.0001491	0.0220
Zbtb10	-0.84	0.0001502	0.0220
Flnb	0.56	0.0001631	0.0238
Uba52	-0.80	0.0001677	0.0243
Chordc1	0.61	0.0001701	0.0245
Uqcrq	0.66	0.0001716	0.0246
Wdr95	3.21	0.0001739	0.0247
Gm6297	1.46	0.0001742	0.0247
Gm10451	3.10	0.0001762	0.0247
Tsc22d1	-0.56	0.0001766	0.0247
Far2	0.79	0.0001814	0.0252
Zfp780b	0.86	0.0001858	0.0257
Pik3c2a	0.69	0.000188	0.0258
Nav2	0.73	0.0001885	0.0258
Frat2	-0.58	0.0001914	0.0259
Hds1	-0.80	0.0001916	0.0259
Dolk	0.96	0.000195	0.0262
Prom1	1.16	0.0001956	0.0262
Zfp760	0.96	0.0002002	0.0266
Bag1	-0.67	0.0002045	0.0270
Chdh	0.56	0.0002056	0.0270
Il18bp	1.04	0.0002085	0.0273
Papss2	0.64	0.0002099	0.0273
Synj1	0.61	0.0002107	0.0273
Megf6	5.64	0.0002132	0.0274
Kcnj2	0.98	0.000217	0.0278
Ptms	-1.12	0.0002187	0.0278
Sgip1	1.39	0.0002276	0.0288
Cd300e	5.49	0.0002295	0.0289
H2-Ab1	0.58	0.0002313	0.0290
Gm10248	-3.82	0.0002337	0.0291
Ubb	-1.54	0.0002374	0.0293
Rps5	-0.73	0.0002376	0.0293
Slc35a5	0.99	0.0002417	0.0296
Zscan29	0.67	0.0002426	0.0296
Cox4i1	-0.88	0.000245	0.0297
Tet2	0.57	0.0002454	0.0297
Vcam1	1.23	0.0002485	0.0298
D230025D16Rik	0.64	0.0002487	0.0298
Sprr1a	6.69	0.0002508	0.0299
Ifitm2	-0.74	0.0002562	0.0304
Itpripl2	0.66	0.0002621	0.0308
B230219D22Rik	0.59	0.0002627	0.0308
Zfp184	-0.82	0.0002699	0.0314
Rc3h2	0.62	0.00027	0.0314
A630072M18Rik	0.85	0.0002786	0.0322
Szt2	0.63	0.0002901	0.0331
E230008N13Rik	-1.87	0.0002912	0.0331
Gsr	0.64	0.0002913	0.0331
Anp32b	-0.60	0.0002921	0.0331
Zdhhc21	0.57	0.000293	0.0331
Stbd1	2.43	0.0002963	0.0333
Serinc4	1.22	0.0002978	0.0333
Rpl9	-0.60	0.000325	0.0362
Dyrk3	-0.68	0.0003265	0.0362
Zfp783	0.86	0.0003307	0.0364
Faim3	5.42	0.0003316	0.0364
Med23	0.61	0.000338	0.0370
Cyp2r1	1.59	0.0003403	0.0371
Gm15915	-0.66	0.0003486	0.0378
Rbm5	0.55	0.0003596	0.0388
Rint1	0.73	0.0003668	0.0394
Stat2	0.75	0.0003733	0.0399
Gbp5	0.92	0.0003755	0.0400
Gas7	0.92	0.0003806	0.0403
Zfp324	0.90	0.0003893	0.0411
2810403A07Rik	0.60	0.000392	0.0412
Rpl36a	-0.57	0.0003952	0.0413
Zbtb40	0.73	0.0004021	0.0419
Slc5a3	0.69	0.0004061	0.0419
Sik1	-0.64	0.0004064	0.0419
Rassf4	0.59	0.0004076	0.0419
Hus1	0.59	0.0004147	0.0425
Mtr	0.58	0.0004174	0.0426
Klf10	-0.71	0.0004232	0.0429
Rab4a	-0.76	0.0004248	0.0429
Tmppe	0.76	0.0004267	0.0429
BC055324	0.63	0.0004277	0.0429
Cybb	0.71	0.0004347	0.0434
Lta4h	0.54	0.0004409	0.0438
Rpl24	-0.57	0.000444	0.0440
Psme2b	1.43	0.0004498	0.0444
Slc7a8	0.60	0.000455	0.0447
Csf2rb2	0.79	0.0004577	0.0448
Hhip	5.66	0.0004609	0.0449
Heatr5b	0.63	0.0004707	0.0457
Met	0.62	0.0004735	0.0458
Rnft1	0.72	0.000477	0.0459
Lig4	1.56	0.0004843	0.0464
Lipo1	0.92	0.0004856	0.0464
Palb2	0.79	0.0004903	0.0465
A530099J19Rik	1.94	0.000492	0.0465
Cr2	1.79	0.0004924	0.0465
Snrnp40	-0.59	0.0005215	0.0487
Ear6	1.83	0.0005219	0.0487
Lsm5	-0.91	0.000522	0.0487
Cdc6	0.59	0.0005255	0.0487
Neat1	0.72	0.000526	0.0487
Pla2g15	1.20	0.0005398	0.0498

Supplemental Table 2. Synergy Genes in dKO LK.

	Relative Expression			
Gene	**miR-146a KO vs WT**	**Tifab KO vs WT**	**dKO vs WT**	**SynergyEffect**
Dgkk	14.84	13.42	52.88	0.53
Ltf	3.64	5.85	16.93	0.56
Ngp	2.17	3.44	9.78	0.57
Cd300e	45.04	12.87	82.53	0.70
Lypd6	15.53	13.63	40.54	0.72
Mir677	14.84	11.06	28.55	0.91
Gpr84	5.64	4.66	10.30	1.00
Timm8a2	20.60	32.50	52.46	1.01
Sfrp1	14.56	30.72	41.74	1.08
Nrg1	5.93	3.57	8.74	1.09
Abca13	2.32	2.92	4.71	1.11
Gdpgp1	2.12	1.54	3.05	1.20
Neo1	10.16	6.63	13.69	1.23
Acpp	2.42	2.91	4.24	1.26
Ncam1	1.64	2.80	3.53	1.26
Spint1	31.74	12.74	32.83	1.35
Vcam1	2.35	3.63	4.33	1.38
Itga1	1.46	1.80	2.30	1.41
Gramd3	1.65	1.68	2.32	1.43
Stc2	5.78	6.02	8.20	1.44
Csf2ra	1.44	1.47	2.01	1.45
Stard5	1.70	1.54	2.22	1.46
Scgb3a1	12.37	21.75	22.95	1.49
Ifi204	2.67	3.12	3.89	1.49
Clca1	1.58	1.69	2.18	1.50
Kcnj2	1.97	1.69	2.39	1.53
Gbp8	9.97	8.33	11.92	1.53
Higd1b	12.37	11.46	15.37	1.55
Mlxipl	18.86	25.93	28.52	1.57
Slfn2	2.76	3.01	3.66	1.58
Papss2	1.56	1.64	2.02	1.58
Clec5a	2.59	2.70	3.22	1.64
Prkcb	2.69	2.21	2.95	1.66
Ceacam1	1.49	1.43	1.75	1.66

Supplemental Table 3. Del(5q) MDS/AML patient characteristics.

Disease	Risk	TIFAB deletion	miR-146a deletion
sAML	1	Yes	No
MDS	0	Yes	No
MDS	0	Yes	No
sAML	1	Yes	Yes
MDS	1	Yes	Yes
MDS	0	Yes	Yes
MDS	1	Yes	Yes
MDS	1	Yes	Yes
MDS	0	Yes	Yes

Risk: 1, high-risk; 0, low-risk
sAML, secondary AML.