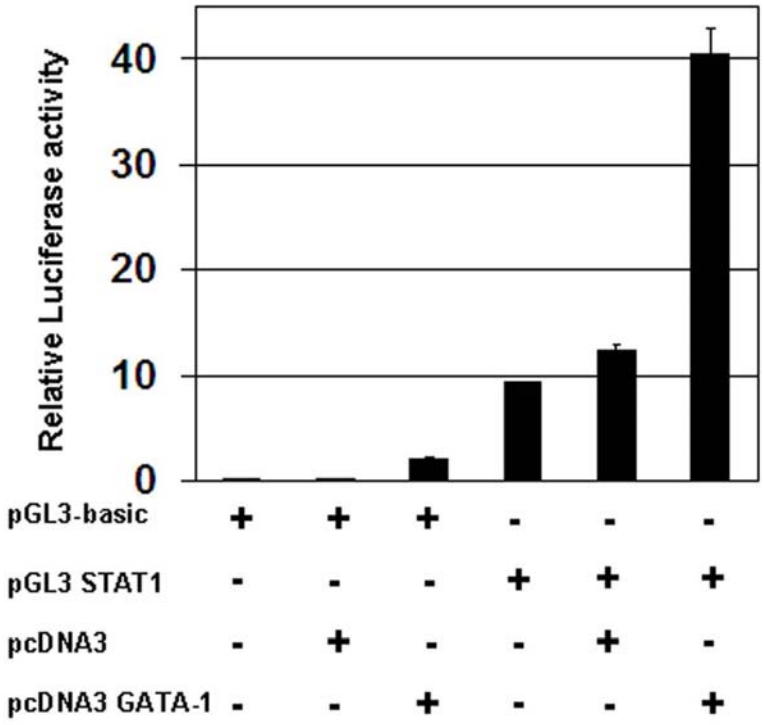
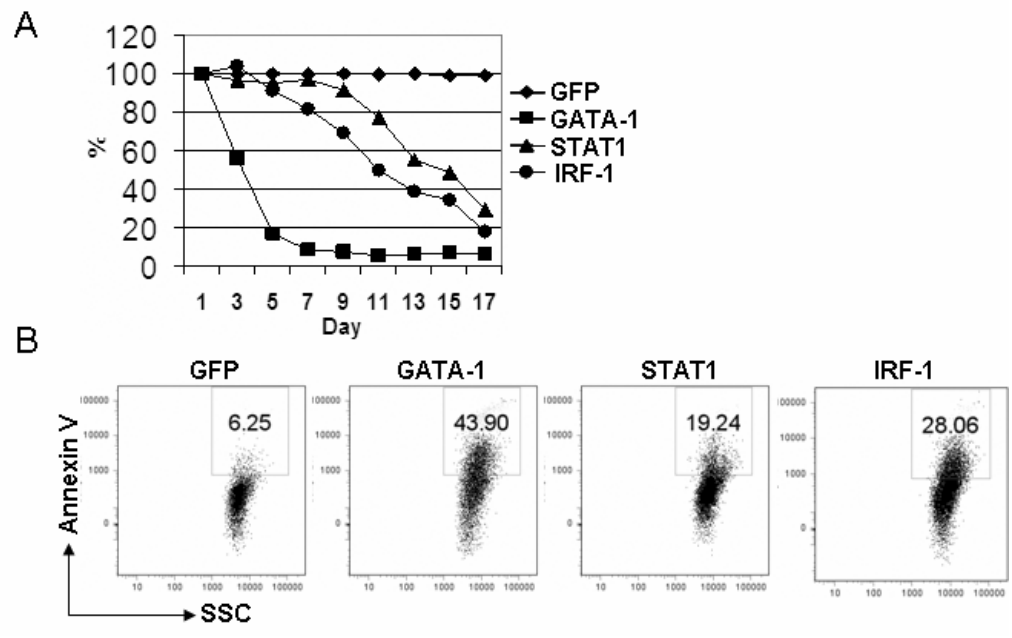
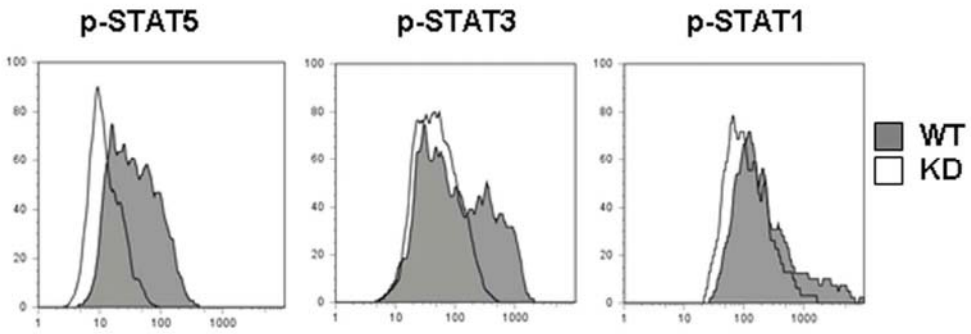


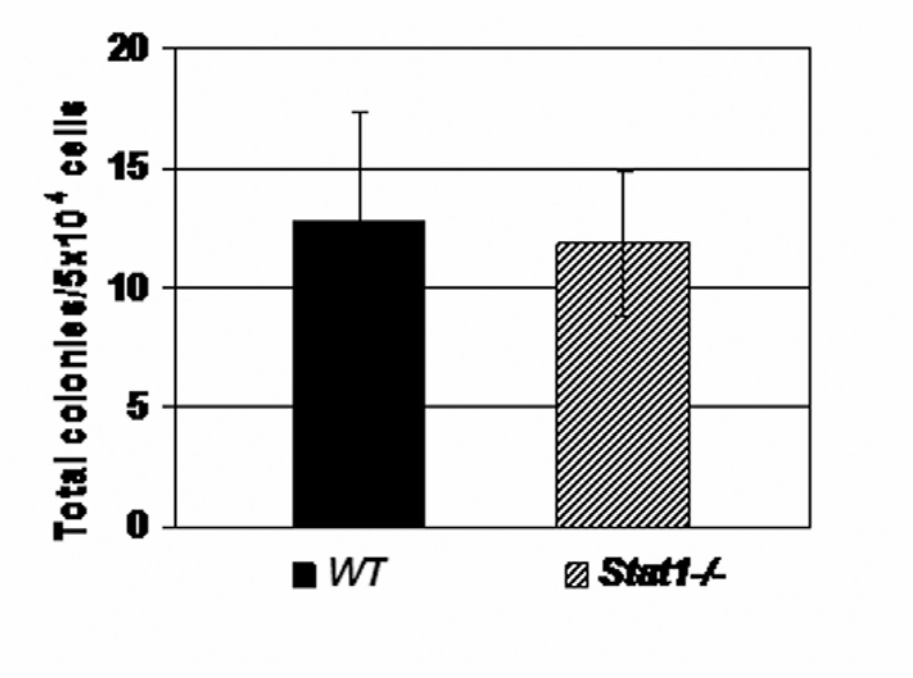
Supplementary Figure 1 GATA-1 can activate the STAT1 promoter. 293 cells were co-transfected with pGL3-basic, pGL3 STAT1, pcDNA3, and/or pcDNA3 GATA-1 as indicated. As an internal control, the same amount of β-galactosidase was included in each transfection. Relative luciferase activity was determined by normalizing the luciferase reading to the β-galactosidase value.



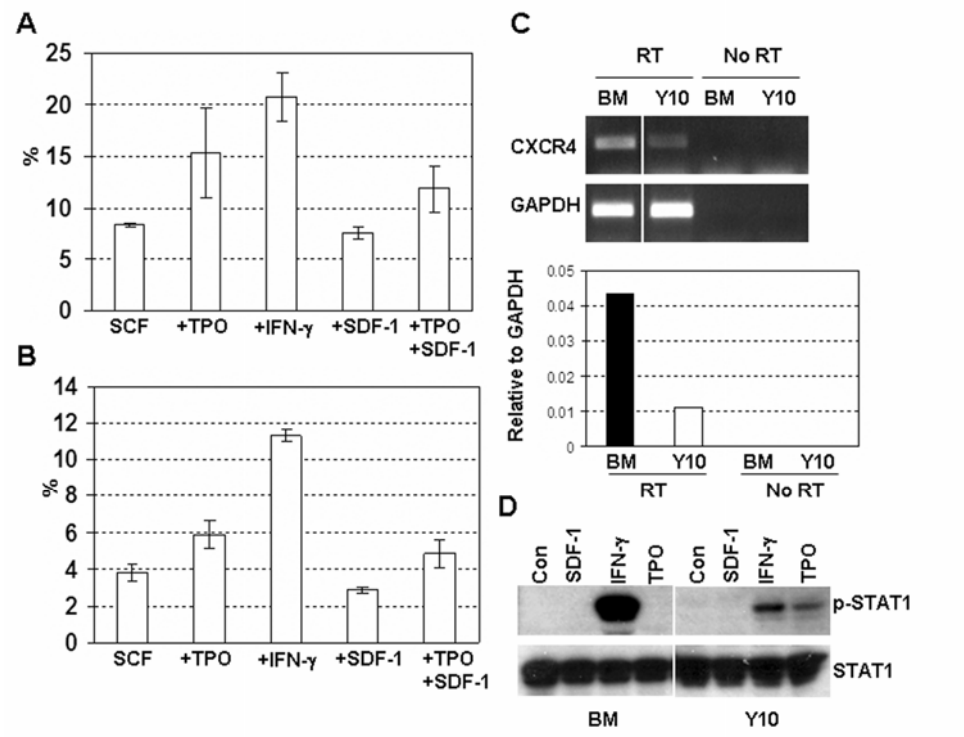
Supplementary Figure 2 Differentiating G1ME cells cease to proliferate and undergo apoptosis. (A) The percentage of GFP, GATA-1, STAT1, or IRF-1 transduced G1ME cells was monitored by flow cytometry over time from the day after the last viral infection (set to day 1 and normalized to 100%) until day 17. (B) The transduced cells were also stained with PE-labeled anti-Annexin V antibody. A gate was set on GFP^+ cells for flow cytometry analysis. Figures are representative of two independent experiments with similar results.



Supplementary Figure 3 High polyploid cells show increased TPO signaling. WT and G1KD megakaryocytes were stained with PE-labeled CD41 antibody and DAPI. Phosphorylation of STAT1, STAT3, and STAT5 in WT and G1KD megakaryocytes were measured by staining Alexa 647-labeled antibodies specific for phosphorylated STAT1, STAT3, and STAT5 respectively. The phosphorylation of STAT1 STAT3, and STAT5 was analyzed in $CD41^+$ cells with high polyploidy (≥8N).



Supplementary Figure 4 CFU-Mk of *Stat1*$^{-/-}$ BM cells. 5×10^4 Bone marrow cells from WT and *Stat1*$^{-/-}$ mice were cultured in MegaCult-C media with cytokines and seeded in chamber slides, following the manufactory instruction (Stem Cell Tech Inc). Megakaryocyte colonies were stained with acetylthiocholiniodide and counted under microscopy. Results were statistics of quadruple from three mice (Mean±SD: 12.75±4.49 for WT; 11.83±3.01 for *Stat1*$^{-/-}$; p=0.56).



Supplementary Figure 5 SDF-1 signaling in megakaryocytes. (A) Lineage-depleted BM cells from WT mice were cultured in the presence of SCF alone, or with TPO (+TPO), with IFN-γ (+IFN-γ), with SDF-1 (+SDF-1), with SDF-1 and TPO (+SDF-1+TPO) for three days. CD41+ cells were measured by flow cytometry. Mean±SD from two independent experiments was shown. (B) $CD42^{+}$ cells were also measured by flow cytometry. (C) CXCR4 mRNA in WT BM and Y10 cells were measured by quantitative RT-PCR. (D) WT BM cells and Y10 cells were stimulated without or with SDF-1 (200 ng/ml), IFN-γ (20 ng/ml), or TPO (20 ng/ml) for 30 minutes. The STAT-1 phosphorylation (p-STAT1) status was assayed by western blot. Note that lanes were run in the same gel in C and D.

Supplementary table 1

Gene detected	Transduced Cell type			
	GFP	GATA-1	STAT1	IRF-1
Gata1	1	190.44±17.89	1.24±0.23	0.81±0.02
Stat1	1	2.14±0.13	16.08±3.04	1.88±0.03
Irf1	1	3.18±0.08	7.43±0.90	131.07±13.02
Gp1ba	1	5.42±0.03	0.98±0.01	0.66±0.05
Gp2ba	1	0.93±0.06	0.85±0.08	0.67±0.21
Nfe2	1	1.30±0.01	1.58±0.28	2.50±0.24
Pf4	1	7.61±0.91	2.64±0.24	1.78±0.06
Tubb1	1	6.56±1.06	0.33±0.003	0.31±0.03
Myc	1	0.56±0.09	0.44±0.11	0.51±0.02
Ets2	1	0.28±0.01	0.36±0.05	0.29±0.01
Gata2	1	0.50±0.03	1.32±0.09	1.27±0.10
Runx1	1	1.90±0.11	1.81±0.30	1.80±0.20
Ccnd1	1	2.95±0.42	18.88±0.79	16.86±4.66
Ccnd2	1	7.73±0.46	11.16±0.03	13.16±2.14
Ccnd3	1	1.14±0.06	0.99±0.002	0.96±0.01
Ccne1	1	2.60±0.29	6.31±1.13	2.94±0.39
Ccne2	1	1.57±0.26	1.38±0.24	0.80±0.08
p16	1	0.71±0.05	1.00±0.25	1.00±0.06
p21	1	0.61±0.04	0.68±0.16	0.63±0.03
p27	1	1.61±0.07	1.89±0.23	1.35±0.67
Bcl2	1	0.42±0.07	1.39±0.14	1.61±0.50
Bclxl	1	0.35±0.04	0.39±0.14	0.33±0.08
Casp1	1	25.52±18.13	67.08±47.06	164.28±76.37
Casp3	1	0.84±0.39	1.56±0.20	1.41±0.34
Casp4	1	72.47±39.27	57.94±31.90	49.34±29.20
Casp9	1	1.51±0.08	2.15±0.33	1.71±0.80
Casp12	1	10.53±4.64	79.79±15.12	385.63±88.70
Mpl	1	0.88±0.052	0.94±0.18	1.09±0.19
Jak2	1	2.13±0.40	2.96±1.08	2.37±0.24
Stat3	1	0.98±0.36	0.71±0.23	0.85±0.08
Stat5a	1	1.67±0.38	1.36±0.16	1.74±0.05
Stat5b	1	1.89±0.41	1.14±0.16	1.13±0.34
Ifngr1	1	3.28±0.61	6.96±0.94	3.50±0.92

Supplementary Material and Methods

Luciferase activity assay

293 cells (1.5×10^5) were seeded in 24-well plate the day before transfection. By using Lipofectamine 2000, cells were transfected with control vector DNA, pGL3 basic, or pGL3 STAT1 promoter, in combination with either pcDNA3 or GATA-1 expression vector pcDNA3 GATA-1. In each transfection, 350 ng of each constructs and 50 ng of β-galactosidase-expressing constructs was used. 48 hr after transfection, cells were lysed in 100 μl PLB (passive lysis buffer). Cell lysates were frozen and thawed once. After centrifuge at 12, 000 rpm 4°C for 2 min, supernatants were collected. Then cell lysates and the substrates of luciferase and β-galactosidase were equilibrated at RT for 30 min. Mixed 100 μl of luciferase or β-galactosidase substrates with 10 μl of cell lysates and read by an Analytical Luminescence Laboratory Monolight 2010. Relative luciferase activity was obtained by normalizing luciferase reading to β-galactosidase readings.

Primer sequences for RT-PCR

Gp2ba	5'-GCCATGAGCTCCAGTCTGAT-3'
	5'-AGGAACAGCACTAGGACCCA-3'
c-Mpl	5'-CTGAGGCATGAACTCCGCTAT-3'
	5'-GTTGGGATGCTGTCGGATGAA-3
Ifngr1	5'-GGAGTGGAGCTTTGACGAG-3'
	5'-AGTCCAGGAACCCGAATACAC-3'
Stat5a	5'-CACTCCTGTACTTGCGAAAG-3'
	5'-CAGGGTTGGGTGGGTACAT-3'
Stat5b	5'-AACAAGCCAGACGGGACCTT-3'
	5'-GTCAGCGAGGGACCGGATA-3'
Stat3	5'-CCCACTCCTTGCCAGTTGTG-3'

	5’-CGCTTGGTGGTGGACGAGAA-3’
Bcl2l11	5’-GGAGGAACCTGAAGATCTGC-3’
	5’-TGCCTTCTCCATACCAGACG-3’
Bclxl	5’-TGTGCGTGGAAAGCGTAGAC-3’
	5’-TGCTGCATTGTTCCCGTAGAG-3’
Casp1	5’-TTGGAGCTCAAGTTGACCTC-3’
	5’-TTCCCTCCTGGATACCATGA-3’
Casp3	5’-CTGACTGGAAAGCCGAAACT-3’
	5’-ATGAACCACGACCCGTCCT-3’
Casp4	5’-GCCAATGGCCGTACACGAA-3’
	5’-ATGCCCTCTGCTGTAAGCTC-3’
Casp9	5’-TGTGTCAAGTTTGCCTACCC-3’
	5’-TTGTAAGTCCCTTTCGCAGAA-3’
Casp12	5’-TGGAGAAGGAGGGACGAAC-3’
	5’-GGCCAGCAAACTGCATTAAC-3’
Runx1	5’-GCACTCTGGTCACCGTCAT-3’
	5’-ATGGTAGGTGGCAACTTGTG-3’
Ccde1	5’-TCCAAGTGGCCTATGTCAAC-3’
	5’-AAGCAGAAGCAGCGAGGAC-3’
Ccne2	5’-TCTCGGAATGTGTAGACTGG-3’
	5’-GCCCTCCTTTTCTGTAGATG-3’
Stat1	5’-AGGGGCCATCACATTCACAT-3’
	5’-AGATACTTCAGGGGATTCTC-3’
Jak2	5’-AGGACAACACTGGCGAGGTG-3’
	5’-GACCCGCACTGTAGCACACT-3’
Tubb1	5’-CTACAATGCCGTGCTATCCA-3’
	5’-AGTGAAGTCGTGATTCCGCT-3’
Gp1ba	5’-GTGCAGAGGGCAAGGCAAGT-3’
	5’-TGACTCAGAGCTGAGGGTCG-3’
Irf1	5’-GGGAAGATAGCCGAAGACCT-3’
	5’-CCTCGAGGGCTGTCAATCT-3’

Nfe2	5’-ACGTGGACATGTACCCAGTGG-3’
	5’-GCCACCTTGTTCTTGCCCCGT-3’
p27	5’-AGTGTCCAGGGATGAGGA-3’
	5’-GGGAACCGGTCTGAAACAT-3’
Ets2	5’-GCAACGTGAATTTGCTCAAC-3’
	5’-GTAGGGACGACTGGCTGTTC-3’
Pf4	5’-AGCATGAGCTCCGCAGCCGGGTTCT-3’
	5’-GTAGGCAGCTAGTAGCTAACTCTCC-3’
Gapdh	5’-AGCCTCGTCCCGTAGACAAA-3’
	5’-CCTTGACTGTGCCGTTGAAT-3’
Cxcr4	5’-CATCTGGACCGCCTTTACC-3’
	5’-TTGAGGGCCTTGCGCTTCT-3’