Supplementary Information

Supplementary Figure 1. Kras G12D mice in the BALB/c background show similar phenotypes as those in the B6 background. Five-seven week old control and Kras G12D mice in the B6 or BALB/c background were injected with pI-pC as described in Materials and Methods. (A) Kaplan-Meier survival curves were plotted against days after the 1st pI-pC injection. Log-rank test was performed to determine statistical significance. (B) Quantitative analysis of HSCs in hind limb bone marrow [BM (H.L.)] and spleen (SP) of control and Kras G12D mice in different backgrounds. HSCs are defined as [CD41 CD48 B220 Gr1 TER119]$^{-}$ CD150^{+} cKit^{+} cells. Data are presented as mean + s.d.. (C,D) Total bone marrow cells were serum- and cytokine- starved for 1 or 2 hours and stimulated with various concentrations of SCF (C) or GM-CSF (D) at 37°C for 10 minutes. Levels of phosphorylated AKT, ERK1/2, and STAT5 were measured using phospho-specific flow cytometry. Non-neutrophil bone marrow cells were gated for data analysis. Myeloid progenitors are enriched in c-Kit^{+} Lin$^{-/low}$ cells, whereas myeloid precursors are enriched in c-Kit^{-} Lin$^{-/low}$ cells. To quantify the activation of AKT, ERK1/2, or STAT5, median intensities of p-AKT, p-ERK, or p-STAT5 at different cytokine concentrations in different groups of animals are compared to those of their respective control cells at 0 ng/ml, which are arbitrarily set at 1. n.s., not significant.

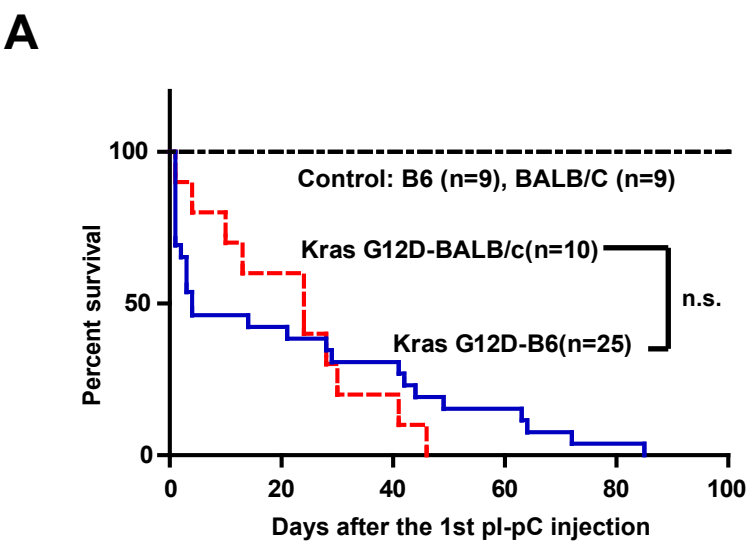
Supplementary Figure 2. Notch1 Type 1 deletions are identified in 100% of Nras G12D/+ or G12D/G12D-induced TALL.

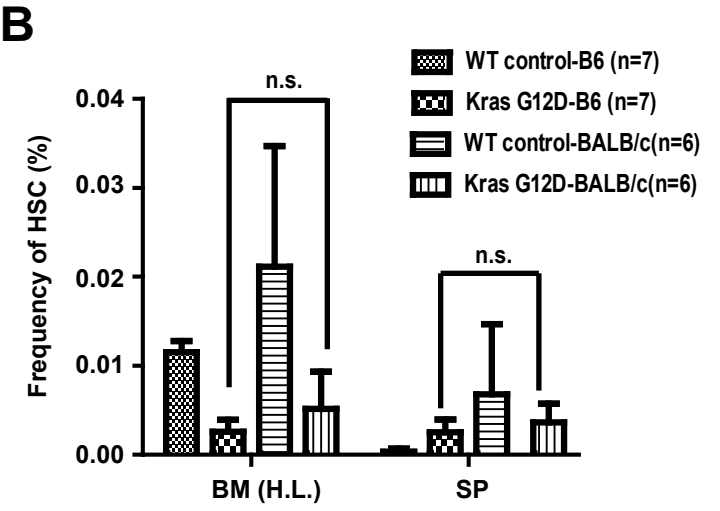
Supplementary Figure 3. Notch1 Type 1 deletions are not detectable in primary Kras G12D bone marrow cells.

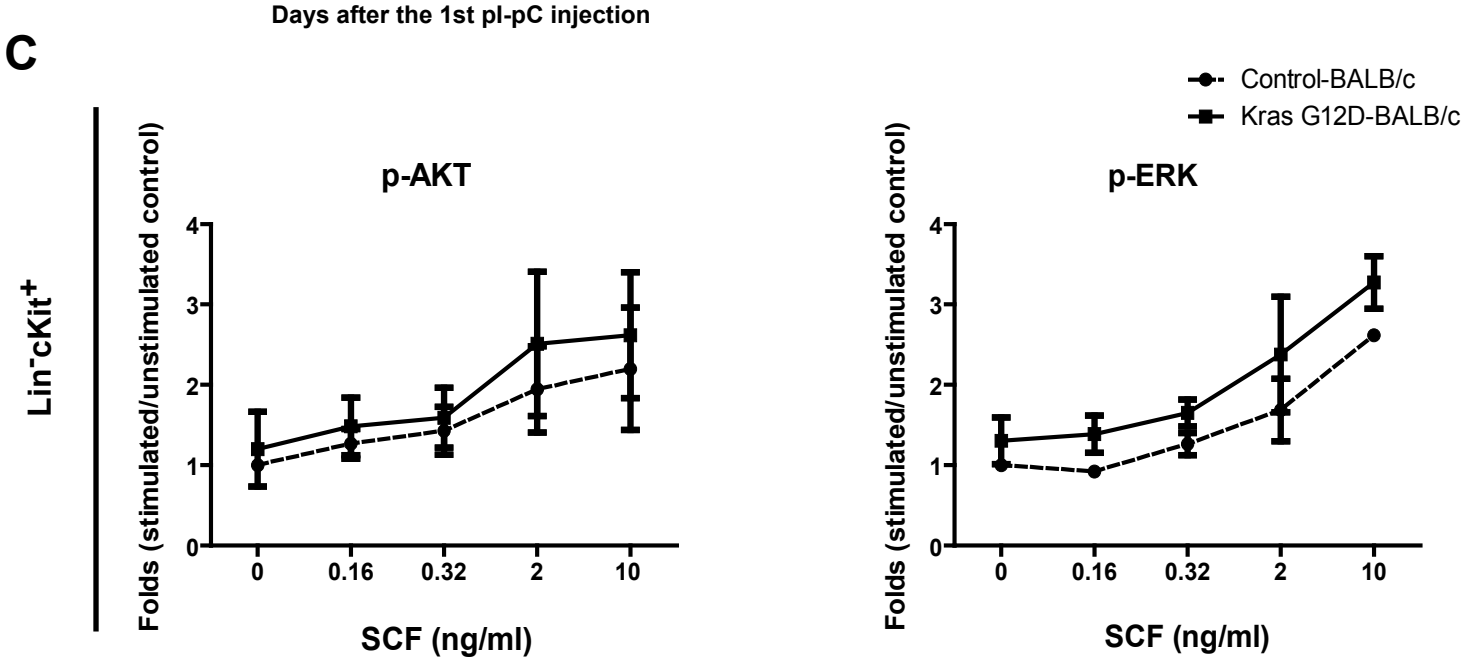
Supplementary Figure 4. The presence of Notch1 Type 1 deletions correlates with T-LIC activity. T-ALL cells were isolated from moribund recipient mice and fractionated into different populations. Sorted cells were further transplanted into 2^{nd} recipients to test for T-LIC activity (A) and presence of Notch1 Type 1 deletion (B).

Supplementary Figure 5. Transplantation of fractionated and serially diluted T-ALL cells. T-ALL was scored for up to 16-20 weeks posttransplant.

Fig S1-Zhang


A
100
50
0
Percent survival
Control: B6 (n=9), BALB/C (n=9)
Kras G12D-BALB/c(n=10)
Kras G12D-B6(n=25)
n.s.
0
20
40
60
80
100
Days after the 1st pI-pC injection


B
WT control-B6 (n=7)
Kras G12D-B6 (n=7)
WT control-BALB/c(n=6)
Kras G12D-BALB/c(n=6)
Frequency of HSC (%)
0.04
0.03
0.02
0.01
0.00
n.s.
n.s.
BM (H.L.)
SP


C
Control-BALB/c
Kras G12D-BALB/c
Lin-cKit+
p-AKT
p-ERK
Folds (stimulated/unstimulated control)
SCF (ng/ml)
0
0.16
0.32
2
10

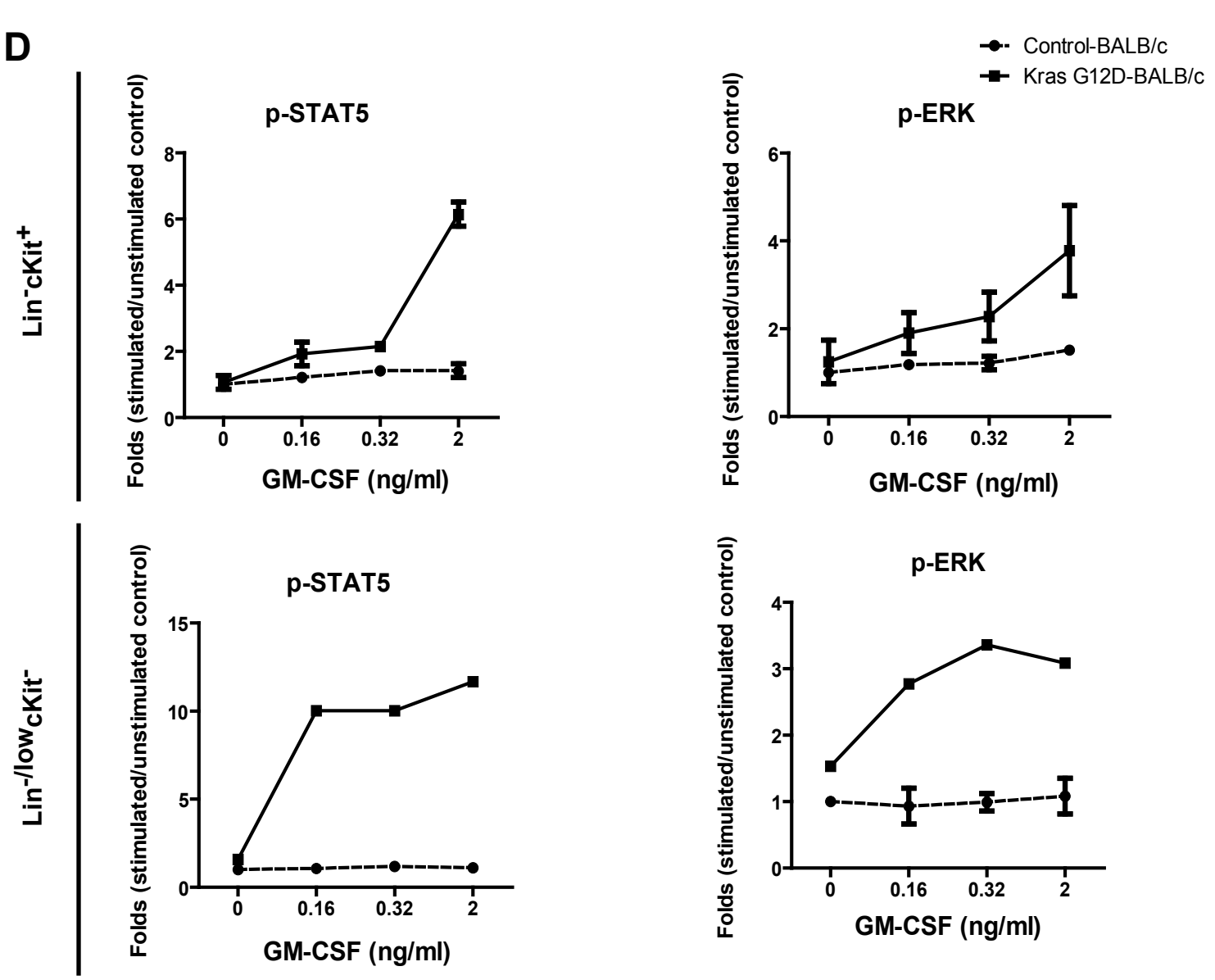

D
Control-BALB/c
Kras G12D-BALB/c
Lin-cKit+
p-STAT5
p-ERK
Folds (stimulated/unstimulated control)
GM-CSF (ng/ml)
Lin-/lowcKit-
0
0.16
0.32
2

Fig S2-Zhang

Fig S3-Zhang

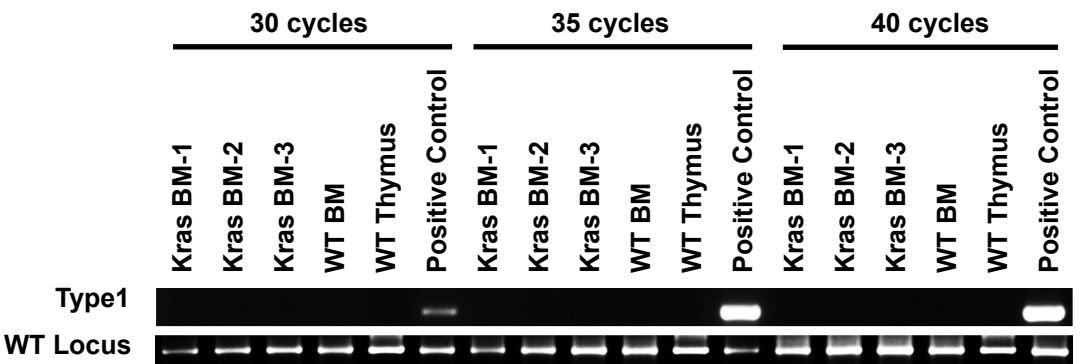

30 cycles
35 cycles
40 cycles
Kras BM-1
Kras BM-2
Kras BM-3
WT BM
WT Thymus
Positive Control
Kras BM-1
Kras BM-2
Kras BM-3
WT BM
WT Thymus
Positive Control
Kras BM-1
Kras BM-2
Kras BM-3
WT BM
WT Thymus
Positive Control
Type1
WT Locus

Fig S4-Zhang

A

Cell Type	Injection dose	# of T-ALL mice/# of transplanted mice
CD8+	10^6	2/2
	10^5	2/2
	10^4	3/3
CD8-	10^5	0/4
	10^4	0/6
	10^3	1/10
	10^2	0/6
	10	0/6

B

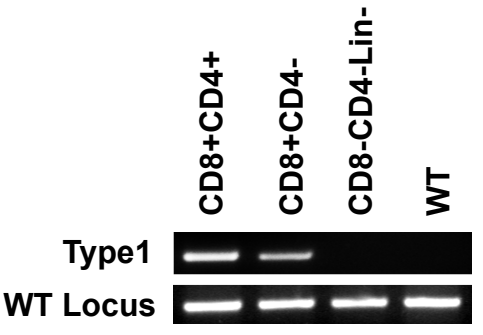
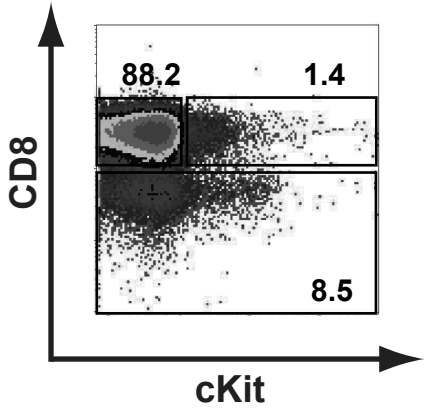


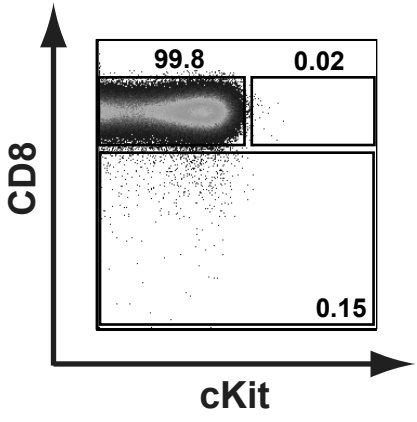
Fig S5-Zhang

Group I



Cell Type	Injection dose	# of T-ALL mice/# of transplanted mice
CD8+ cKit-	10^6	0/6
	10^5	0/4
CD8+cKit+	10^5	2/2
	10^4	2/2
	10^3	1/2
	10^2	0/2

Group II



Cell Type	Injection dose	# of T-ALL mice/# of transplanted mice
CD8+ cKit-	10^6	1/1
	10^5	2/2
	10^4	3/3
	10^3	2/2
CD8+cKit+	10^3	2/6
	10^2	0/8