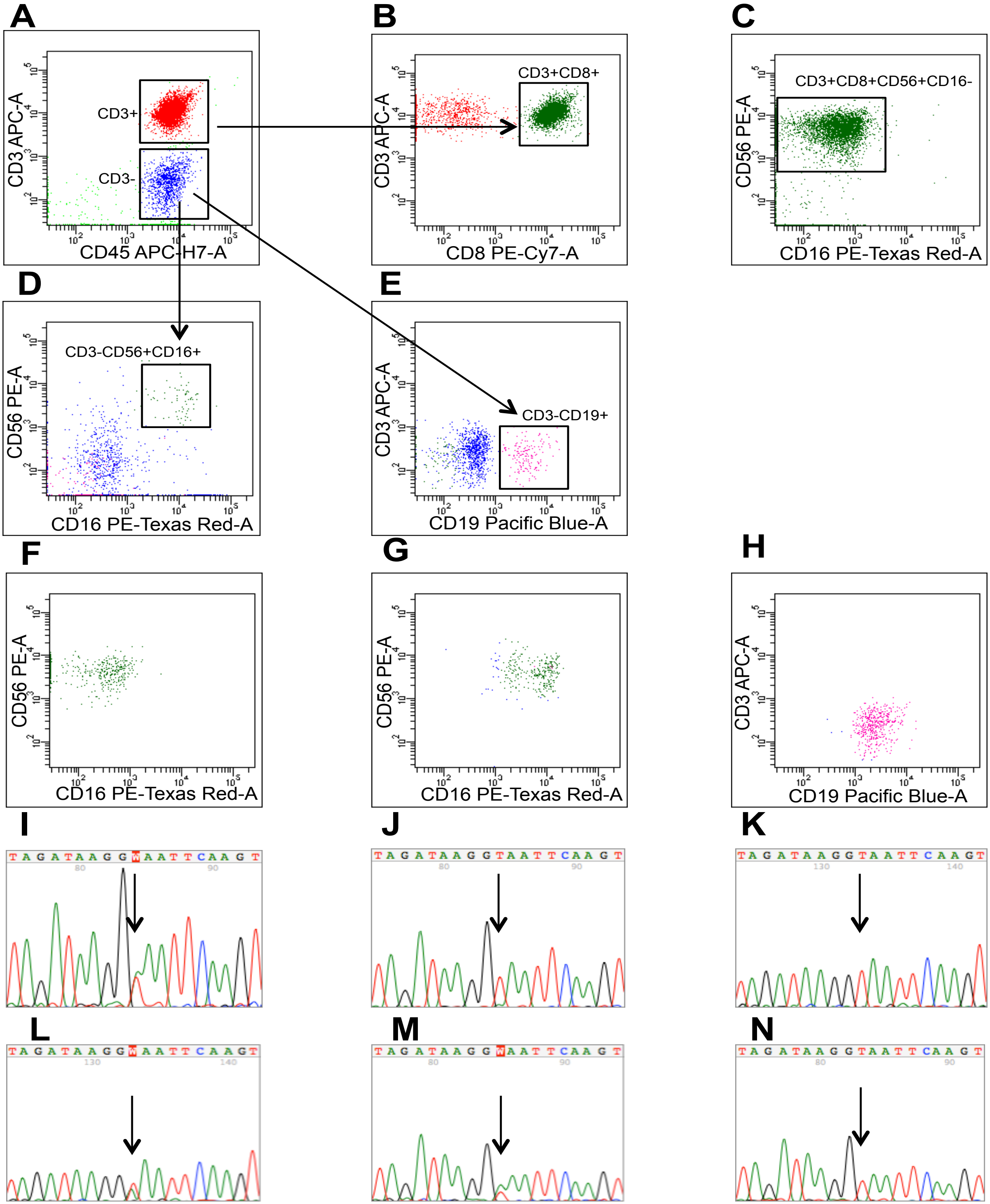
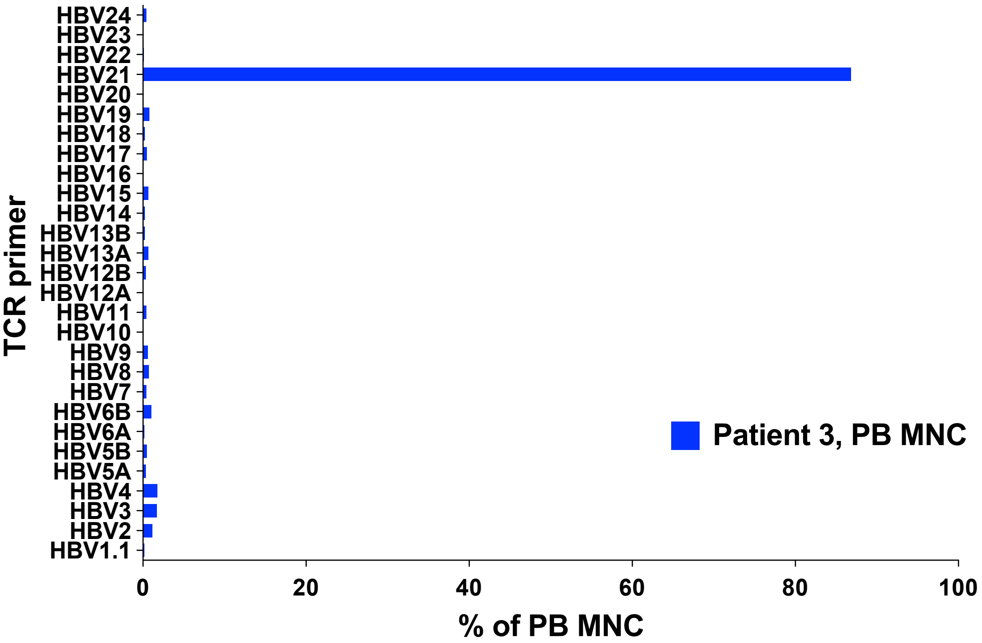
Supplementary Figure 1

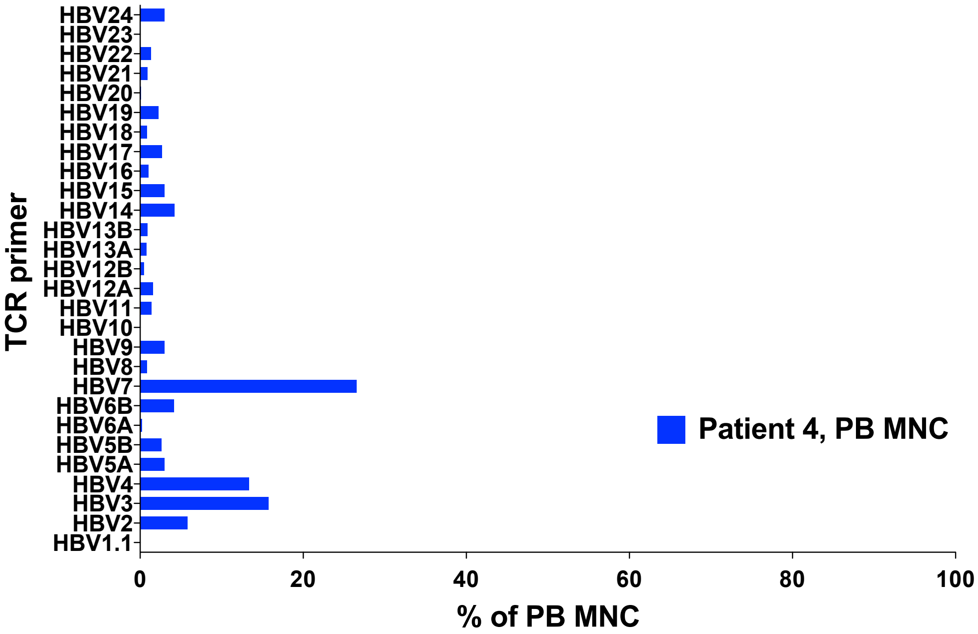


Supplementary Figure 1. STAT5b sequencing from sorted fractions of patient 1 and STAT5b sequencing results of patient 1 and 2.

CD3+CD8+CD56+ leukemic LGLs (A-C), CD3^{neg}CD16+CD56+NK-cells (D), and CD3^{neg}CD19+ B-cells (E) were sorted from patient 1 with STAT5b mutation Y665F. Approximately 83% of patient 1's CD3+CD8+ cells expressed also CD56, but not CD16. Panels F-G represent the purity of sorted fractions: leukemic LGLs (F), NK-cells (G), and B-cells (H). STAT5b mutation Y665F is seen in CD8+CD56+ cells DNA (I), but not in NK-cells (G) or B-cells (H). Similar gating and sorting strategy was done with patient 2's sample (flow cytometry results not shown). Mutation Y665F is visible in CD3+CD8+CD56+ DNA (L), and surprisingly, also in CD3^{neg}CD16+CD56+ NK-cell DNA (M). B-cells show only wild-type allele (N).

Supplementary Figure 2

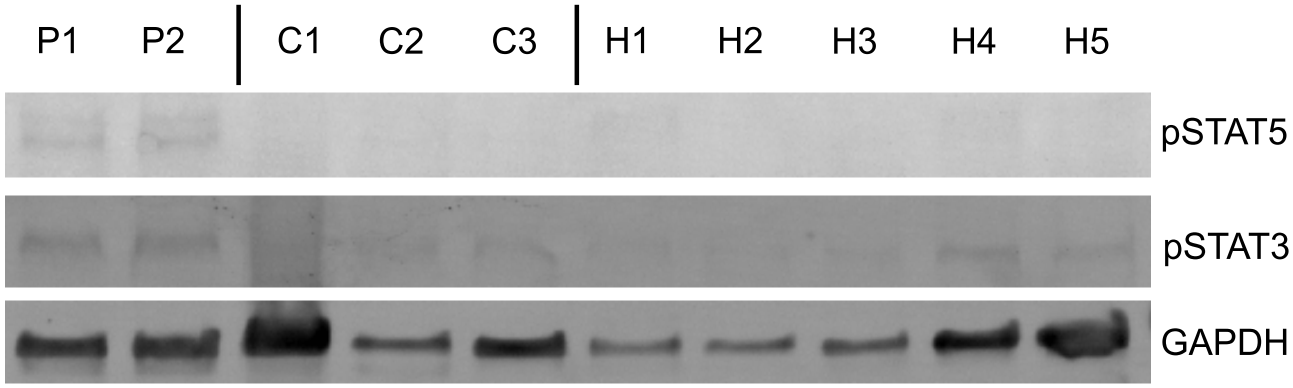




Supplementary Figure 2. Patients 3 and 4 with STAT5b mutation N642H: T-cell receptor rearrangement RT-PCR results.

T-cell receptor rearrangement and T-cell clonality in patients 3 and 4 was analyzed using RT-PCR (the method described by Ochsenreither et al. J Transl Med 2008). RNA was derived from PB MNCs of patients 3 and 4. Samples were analyzed with SYBR Green RT-PCR (Life Technologies). The method is semi-quantitive and it can be used to determine clonality in given sample. In patient 3 with CD56+ T-cell LGL leukemia, 86% of PB MNC consisted of a single clone (HBV21). In patient 4 with NK-cell LGL leukemia, there was one expansion of 27% (the average TCR-percent across all probes was 3.6% and a single standard deviation was 5.8%, thus HBV7 is +3SD above average representing abnormal expansion).

Supplementary Figure 3



Supplementary Figure 3. Phospho-STAT3 and phospho-STAT5 western blotting results. Freshly isolated PB MNC's were sheared into cytosolic and nuclear fractions. We analyzed pSTAT3 and pSTAT5 expression in nuclear fractions using samples from 2 LGL-leukemia patients with STAT5b mutation Y665F (Patients 1-2, Table 1), 3 control LGL-leukemia patients with wild-type STAT3/STAT5b (C1-3) and 5 healthy controls (H1-5). The patients with STAT5b mutation expressed pSTAT5 and to a lesser extent pSTAT3 in nuclear lysates. Other LGL patients or healthy controls did not express pSTAT5.

Supplementary Table 1. Patient cohort characteristics

	Median age at diagnosis, years (range)	Males	Treatment *	Hb, g/l median (range)	WBC, 10^6/l median (range)	Lymph, 10^6/l median (range)
T-LGL (n=173)	65 (17-89)	103/173 (60%)	115/173 (66%)	118 (59-172)	6.1 (0.8-90)	3.4 (0.4-85.5)
NK-LGL (n=38)	60 (27-77)	25/38 (66%)	18/38 (47%)	129 (58-168)	9.9 (1.8-164.7)	6.8 (1.0-131.8)
Total (n=211)	64 (17-89)	128/211 (61%)	133/211 (63%)	121 (58-172)	6.5 (0.8-164.7)	3.7 (0.4-131.8)

*Treatment at any timepoint
Abbreviations: Hb, hemoglobin; WBC, white blood cell count; Lymph, lymphocytes

Supplementary Table 2. Capillary sequencing primers

Primer name	**Primer sequence**
STAT5a_exon15_16_F	5’- GCCCTGACTCGGGGGTTCCT-3’
STAT5a_exon15_16_R	5’- GTGGCGGGGCAAGGGAACAA-3’
STAT5a_exon_18_19_F	5’- ACATGGGGCGTGGGCTTCCA-3’
STAT5a_exon_18_19_R	5’- GCCAGCCCTCCAGGAGTCCA-3’
STAT5b_exon_14_15_F	5’- AGGCGGGGATATTTGTATGCCTCT-3’
STAT5b_exon_14_15_R	5’- TGTTCATGTGTAACCATGCTGCCAT-3’
STAT5b_exon_17_18_F	5’- AGGTGGTTGTGTTCTGTCCT-3’
STAT5b_exon_17_18_R	5’- CAGTTCCTCCCCTGTGGAC-3’
BMPR2_F	5’- GGGAGTGTCTCCCAGAATTTGGCT-3’
BMPR2_R	5’- ACGGCAAACTTACCTCCAACAGTTT-3’
EBF3_F	5’- CACCGCGCTGCTTCGAGCTA-3’
EBF3_R	5’- GCGCAGAAAAGGAGAGCCAGC-3’
SGMS2_F	5’-TTTCATTAGGAGTAAAACCAACCA-3’
SGMS2_R	5’-CATTGTAATGCAGCGATACAGG-3’

Supplementary Table 3. Locus specific primer sequences carrying tails corresponding to the Illumina adapter sequences. Locus specific primer sequences are underlined.

Primer name	**Primer sequence**
STAT5B_exon16-F	5’-ACACTCTTTCCCTACACGACGCTCTTCCGATCTTGTTGGGGTTTTAAGATTTCC-3’
STAT5B_exon16-R	5’-AGACGTGTGCTCTTCCGATCTCAAATCAGAATGCGAACATTG-3’
STAT5A_exon17-F	5’-ACACTCTTTCCCTACACGACGCTCTTCCGATCTTCCTGCTGCTGGTGGATTAT-3’
STAT5A_exon17-R	5'-AGACGTGTGCTCTTCCGATCTAGCCCAAGGCTTTGTCTATG-3'

Supplementary Table 4. Illumina TruSeq Adapter primer sequences. TruSeq Adapter index primers (indices 1-48) are adapted from Illumina, rest of the index primers (FIMM 61-100) contain non-Illumina indices. Index sequences are underlined.

Primer name	Primer sequence
TruSeq Universal Adapter	5’-AATGATACGGCGACCACCGAGATCTACACTCTTTCCCTACACGACGCTCTTCCGATC*T-3’
TruSeq Adapter index 1	5'-CAAGCAGAAGACGGCATACGAGATCGTGATGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 2	5'-CAAGCAGAAGACGGCATACGAGATACATCGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 3	5'-CAAGCAGAAGACGGCATACGAGATGCCTAAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 4	5'-CAAGCAGAAGACGGCATACGAGATTGGTCAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 5	5'-CAAGCAGAAGACGGCATACGAGATCACTGTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 6	5'-CAAGCAGAAGACGGCATACGAGATATTGGCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 7	5'-CAAGCAGAAGACGGCATACGAGATGATCTGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 8	5'-CAAGCAGAAGACGGCATACGAGATTCAAGTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 9	5'-CAAGCAGAAGACGGCATACGAGATCTGATCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 10	5'-CAAGCAGAAGACGGCATACGAGATAAGCTAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 11	5'-CAAGCAGAAGACGGCATACGAGATGTAGCCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 12	5'-CAAGCAGAAGACGGCATACGAGATTACAAGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 13	5'-CAAGCAGAAGACGGCATACGAGATTTGACTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'

TruSeq Adapter index 14	5'-CAAGCAGAAGACGGCATACGAGATGGAACTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 15	5'-CAAGCAGAAGACGGCATACGAGATTGACATGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 16	5'-CAAGCAGAAGACGGCATACGAGATGGACGGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 17	5'-CAAGCAGAAGACGGCATACGAGATCTCTACGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 18	5'-CAAGCAGAAGACGGCATACGAGATGCGGACGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 19	5'-CAAGCAGAAGACGGCATACGAGATTTTCACGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 20	5'-CAAGCAGAAGACGGCATACGAGATGGCCACGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 21	5'-CAAGCAGAAGACGGCATACGAGATCGAAACGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 22	5'-CAAGCAGAAGACGGCATACGAGATCGTACGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 23	5'-CAAGCAGAAGACGGCATACGAGATCCACTCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 24	5'-CAAGCAGAAGACGGCATACGAGATGCTACCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 25	5'-CAAGCAGAAGACGGCATACGAGATAGCTAGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 26	5'-CAAGCAGAAGACGGCATACGAGATGCTCATGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 27	5'-CAAGCAGAAGACGGCATACGAGATGTATAGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 28	5'-CAAGCAGAAGACGGCATACGAGATTCTGAGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 29	5'-CAAGCAGAAGACGGCATACGAGATCAGCAGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 30	5'-CAAGCAGAAGACGGCATACGAGATCGATTAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'

TruSeq Adapter index 31	5'-CAAGCAGAAGACGGCATACGAGATGCTGTAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 32	5'-CAAGCAGAAGACGGCATACGAGATATTATAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 33	5'-CAAGCAGAAGACGGCATACGAGATGAATGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 34	5'-CAAGCAGAAGACGGCATACGAGATTCGGGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 35	5'-CAAGCAGAAGACGGCATACGAGATCTTCGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 36	5'-CAAGCAGAAGACGGCATACGAGATTGCCGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 37	5'-CAAGCAGAAGACGGCATACGAGATCGCGGCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 38	5'-CAAGCAGAAGACGGCATACGAGATTAAGGCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 39	5'-CAAGCAGAAGACGGCATACGAGATTGGAGCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 40	5'-CAAGCAGAAGACGGCATACGAGATTGTGCCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 41	5'-CAAGCAGAAGACGGCATACGAGATCAGGCCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 42	5'-CAAGCAGAAGACGGCATACGAGATGGTAGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 43	5'-CAAGCAGAAGACGGCATACGAGATCATTCAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 44	5'-CAAGCAGAAGACGGCATACGAGATATGGCAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 45	5'-CAAGCAGAAGACGGCATACGAGATCCAGCAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 46	5'-CAAGCAGAAGACGGCATACGAGATGCGCCAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter index 47	5'-CAAGCAGAAGACGGCATACGAGATTTCGAAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'

TruSeq Adapter index 48	5'-CAAGCAGAAGACGGCATACGAGATGGAGAAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 61	5'-CAAGCAGAAGACGGCATACGAGATAACGATGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 62	5'-CAAGCAGAAGACGGCATACGAGATAATCGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 63	5'-CAAGCAGAAGACGGCATACGAGATACACTTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 64	5'-CAAGCAGAAGACGGCATACGAGATACCACCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 65	5'-CAAGCAGAAGACGGCATACGAGATACCTTAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 66	5'-CAAGCAGAAGACGGCATACGAGATACGCGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 67	5'-CAAGCAGAAGACGGCATACGAGATACTAGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 68	5'-CAAGCAGAAGACGGCATACGAGATAGAACTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 69	5'-CAAGCAGAAGACGGCATACGAGATAGACTAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 70	5'-CAAGCAGAAGACGGCATACGAGATAGCAATGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 71	5'-CAAGCAGAAGACGGCATACGAGATAGCGACGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 72	5'-CAAGCAGAAGACGGCATACGAGATAGGATTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 73	5'-CAAGCAGAAGACGGCATACGAGATAGTAGTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 74	5'-CAAGCAGAAGACGGCATACGAGATAGTGCGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 75	5'-CAAGCAGAAGACGGCATACGAGATATAACGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 76	5'-CAAGCAGAAGACGGCATACGAGATATAGCAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'

TruSeq Adapter FIMM 77	5'-CAAGCAGAAGACGGCATACGAGATATCTTGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 78	5'-CAAGCAGAAGACGGCATACGAGATATGATGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 79	5'-CAAGCAGAAGACGGCATACGAGATCAAGGCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 80	5'-CAAGCAGAAGACGGCATACGAGATCAGACGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 81	5'-CAAGCAGAAGACGGCATACGAGATCAGCGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 82	5'-CAAGCAGAAGACGGCATACGAGATCATGAAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 83	5'-CAAGCAGAAGACGGCATACGAGATCCAGCTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 84	5'-CAAGCAGAAGACGGCATACGAGATCCTAGCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 85	5'-CAAGCAGAAGACGGCATACGAGATCGATAAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 86	5'-CAAGCAGAAGACGGCATACGAGATCGCATCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 87	5'-CAAGCAGAAGACGGCATACGAGATCGGATGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 88	5'-CAAGCAGAAGACGGCATACGAGATCGTGTCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 89	5'-CAAGCAGAAGACGGCATACGAGATCTACCGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 90	5'-CAAGCAGAAGACGGCATACGAGATCTCCAGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 91	5'-CAAGCAGAAGACGGCATACGAGATCTGAGGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 92	5'-CAAGCAGAAGACGGCATACGAGATCTTAGTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 93	5'-CAAGCAGAAGACGGCATACGAGATGAACTAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'

TruSeq Adapter FIMM 94	5'-CAAGCAGAAGACGGCATACGAGATGAATTGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 95	5'-CAAGCAGAAGACGGCATACGAGATGAGATTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 96	5'-CAAGCAGAAGACGGCATACGAGATGCAGTGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 97	5'-CAAGCAGAAGACGGCATACGAGATGCGTCTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 98	5'-CAAGCAGAAGACGGCATACGAGATGCTGTCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 99	5'-CAAGCAGAAGACGGCATACGAGATGGAGTCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'
TruSeq Adapter FIMM 100	5'-CAAGCAGAAGACGGCATACGAGATGTATGAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATC*T-3'

---

* denotes a phosphorothioate-modified bond

[1] Oligonucleotide sequences © 200-2012 Illumina, Inc. All rights reserved. Derivative works created by Illumina customers are authorized for use with Illumina instruments and products only. All other uses are strictly prohibited.

Supplementary Table 5. STAT5b mutagenesis primers

Primer name	**Primer sequence**
STAT5b_mutagenesis_Y665F_F	5'- GAG ACT TGA ATT TCC TTA TCT ACG TGT TTC -3'
STAT5b_mutagenesis_Y665F_R	5'- GAA ACA CGT AGA TAA GGA AAT TCA AGT CTC -3'
STAT5b_mutagenesis_N642H_F	5'- GAA AGA ATG TTT TGG CAT CTG ATG CCT TTT AC -3'
STAT5b_mutagenesis_N642H_R	5'- GTA AAA GGC ATC AGA TGC CAA AAC ATT CTT TC -3'
STAT5b_mutagenesis_sequencing_F1	5'- GGACTTTCCAAAATGTCG-3'
STAT5b_mutagenesis_sequencing_F2	5'- GAACAGAGGTTGGTCCGAGA-3'
STAT5b_mutagenesis_sequencing_F3	5'- GTCACGCAGGACACAGAGAA-3'
STAT5b_mutagenesis_sequencing_F4	5'- GAACACCCGCAATGATTACA-3'
STAT5b_mutagenesis_sequencing_F5	5'- CAACAGGCCCATGACCTACT-3'
STAT5b_mutagenesis_sequencing_R	5'- CCACCAGCCTTGTCCTAAT-3'

Supplementary Table 6. Mutations in *STAT5b* detected by Amplicon sequencing

No	Gene	Chr	Position	Mut. type	Var. base	Ref. base	Var. Freq (%)*	Coverage **	Protein
3	STAT5b	17	40359729	Missense, homozygous	G	T	94	480	N642H
4	STAT5b	17	40359729	Missense, heterozygous	G	T	48	942	N642H

Abbreviations: No, number; Chr, chromosome; Var_base, variant base; Ref_base, reference base; Var. Freq, variant frequency

*The percentage of variant bases at the site of the mutation

** The number of reads aligned to the reference genome at the site of the mutation

Supplementary Table 7. Gene expression levels in LGL leukemia patients with STAT5b Y665F mutation versus healthy control CD8+ RNA. The top genes with expression fold change over 2.5 or under 2.5 ($\log_{10}$-scale) are displayed.

Gene	Description	P-value	Fold change ($\log_{10}$)
CX3CR1	chemokine (C-X3-C motif) receptor 1	0,005588	4,965
PRF1	perforin 1 (pore forming protein)	0,004729	3,41
GIMAP4	GTPase, IMAP family member 4	0,003885	3,375
SLC1A7	solute carrier family 1 (glutamate transporter), member 7	0,01883	3,16
LAIR2	leukocyte-associated immunoglobulin-like receptor 2	0,00749	3,075
FGR	Gardner-Rasheed feline sarcoma viral (v-fgr) oncogene homolog	0,003885	3,055
RAP2A	RAP2A, member of RAS oncogene family	0,010534	3,02
PCMT1	protein-L-isoaspartate (D-aspartate) O-methyltransferase	0,003885	2,93
RAB10	RAB10, member RAS oncogene family	0,003885	2,9
PRR5L	proline rich 5 like	0,004098	2,89
BATF	basic leucine zipper transcription factor, ATF-like	0,020192	2,84
DCAF7	DDB1 and CUL4 associated factor 7	0,003885	2,825
AKR1C3	aldo-keto reductase family 1, member C3 (3-alpha hydroxysteroid dehydrogenase, type II)	0,005109	2,81
PTGER2	prostaglandin E receptor 2 (subtype EP2), 53kDa	0,005702	2,805
PLEK	pleckstrin	0,003909	2,8
FGL2	fibrinogen-like 2	0,015298	2,795
LOC100130633	hypothetical protein LOC100130633	0,003982	2,785
B4GALT5	UDP-Gal:betaGlcNAc beta 1,4- galactosyltransferase, polypeptide 5	0,008053	2,745
KLRD1	killer cell lectin-like receptor subfamily D, member 1	0,005251	2,735
ERBB2	v-erb-b2 erythroblastic leukemia viral oncogene homolog 2, neuro/glioblastoma derived oncogene homolog (avian)	0,027067	2,675
ITGB1	integrin, beta 1 (fibronectin receptor, beta polypeptide, antigen CD29 includes MDF2, MSK12)	0,005244	2,675
KLRG1	killer cell lectin-like receptor subfamily G, member 1	0,020797	2,65
PTGDR	prostaglandin D2 receptor (DP)	0,009879	2,64
PLEKHG3	pleckstrin homology domain containing, family G (with RhoGef domain) member 3	0,006436	2,625
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	0,021429	2,61
APOBEC3G	apolipoprotein B mRNA editing enzyme, catalytic polypeptide-like 3G	0,004159	2,57
PYHIN1	pyrin and HIN domain family, member 1	0,021803	2,555
KLRF1	killer cell lectin-like receptor subfamily F, member 1	0,003885	2,555
CCDC56	coiled-coil domain containing 56	0,003885	2,53
LTB	lymphotoxin beta (TNF superfamily, member 3)	0,014099	-2,51
TRAF3IP2	TRAF3 interacting protein 2	0,003885	-2,51
MAFF	v-maf musculoaponeurotic fibrosarcoma oncogene homolog F (avian)	0,01022	-2,52
TSPYL2	TSPY-like 2	0,007409	-2,56
PASK	PAS domain containing serine/threonine kinase	0,006749	-2,585
CD83	CD83 molecule	0,003885	-2,595
ACTN1	actinin, alpha 1	0,035869	-2,615
CENPV	centromere protein V	0,005118	-2,615
NR4A2	nuclear receptor subfamily 4, group A, member 2	0,028072	-2,625

LEF1	lymphoid enhancer-binding factor 1	0,016946	-2,66
SGK1	serum/glucocorticoid regulated kinase 1	0,029676	-2,72
PDE4B	phosphodiesterase 4B, cAMP-specific	0,020235	-2,78
INF2	inverted formin, FH2 and WH2 domain containing	0,008284	-2,8
SPINT2	serine peptidase inhibitor, Kunitz type, 2	0,003924	-2,84
TNF	tumor necrosis factor	0,003885	-2,925
SIK1	salt-inducible kinase 1	0,013065	-2,96
TNFAIP3	tumor necrosis factor, alpha-induced protein 3	0,004795	-2,97
IRS2	insulin receptor substrate 2	0,004962	-2,995
SPRY1	sprouty homolog 1, antagonist of FGF signaling (Drosophila)	0,003885	-3,005
CSRNP1	cysteine-serine-rich nuclear protein 1	0,048419	-3,025
TCEA3	transcription elongation factor A (SII), 3	0,005611	-3,045
RGS1	regulator of G-protein signaling 1	0,006621	-3,305
KLRB1	killer cell lectin-like receptor subfamily B, member 1	0,003885	-3,34
NELL2	NEL-like 2 (chicken)	0,00828	-3,415
CCR7	chemokine (C-C motif) receptor 7	0,01742	-3,44
MAL	mal, T-cell differentiation protein	0,003924	-3,48
IL8	interleukin 8	0,013286	-3,675

Supplementary Table 8. Over-represented signaling pathways (KEGG database) in *STAT5b* Y665F mutated patients compared to healthy control CD8+ RNA

KEGG - test for over-representation

	ID	Pvalue	OddsRatio	ExpCount	Count	Size	Term
1	03010	0	4.87	17.93	48	88	Ribosome
2	04141	0	2.47	31.18	58	153	Protein processing in endoplasmic reticulum
3	05131	2e-06	3.61	12.43	29	61	Shigellosis
4	03040	2e-06	2.57	24.05	46	118	Spliceosome
5	03020	9.1e-05	5.03	5.09	14	25	RNA polymerase
6	04660	0.000187	2.24	20.38	36	100	T cell receptor signaling pathway
7	00510	0.000215	3.17	9.17	20	45	N-Glycan biosynthesis
8	03050	0.000309	3.04	9.37	20	46	Proteasome
9	05220	0.000378	2.52	13.65	26	67	Chronic myeloid leukemia
10	04110	0.00046	1.99	25.07	41	123	Cell cycle
11	04623	0.000939	2.78	9.37	19	46	Cytosolic DNA-sensing pathway
12	05212	0.001115	2.38	13.04	24	64	Pancreatic cancer
13	00071	0.00131	3.01	7.54	16	37	Fatty acid metabolism
14	04710	0.001824	3.94	4.48	11	22	Circadian rhythm - mammal
15	00520	0.002241	2.68	8.56	17	42	Amino sugar and nucleotide sugar metabolism
16	05110	0.00259	2.39	10.8	20	53	Vibrio cholerae infection
17	03018	0.002875	2.31	11.62	21	57	RNA degradation
18	01100	0.003068	1.26	209.5	242	1028	Metabolic pathways
19	00280	0.003453	2.63	8.15	16	40	Valine, leucine and isoleucine degradation
20	04722	0.003463	1.79	24.25	37	119	Neurotrophin signaling pathway
21	04650	0.003463	1.79	24.25	37	119	Natural killer cell mediated cytotoxicity
22	04662	0.005208	2.07	13.65	23	67	B cell receptor signaling pathway
23	00670	0.007789	3.93	3.26	8	16	One carbon pool by folate
24	05223	0.009291	2.15	10.39	18	51	Non-small cell lung cancer
25	00062	0.011242	6.54	1.63	5	8	Fatty acid elongation in mitochondria
26	00240	0.011879	1.76	18.54	28	91	Pyrimidine metabolism
27	00230	0.01261	1.55	31.18	43	153	Purine metabolism
28	05120	0.012993	1.93	13.04	21	64	Epithelial cell signaling in Helicobacter pylori infection
29	05222	0.01369	1.82	15.49	24	76	Small cell lung cancer
30	05215	0.013861	1.77	17.12	26	84	Prostate cancer
31	05100	0.015872	1.85	14.06	22	69	Bacterial invasion of epithelial cells
32	05213	0.016808	2.03	10.19	17	50	Endometrial cancer
33	05142	0.021662	1.67	18.54	27	91	Chagas disease
34	04520	0.022213	1.74	15.28	23	75	Adherens junction
35	03420	0.023357	2.04	8.97	15	44	Nucleotide excision repair
36	00563	0.030308	2.52	4.69	9	23	Glycosylphosphatidylinositol(GPI)-anchor biosynthesis
37	05214	0.035864	1.74	12.64	19	62	Glioma
38	04130	0.038647	2.05	7.13	12	35	SNARE interactions in vesicular transport
39	04621	0.042224	1.76	11.21	17	55	NOD-like receptor signaling pathway
40	00190	0.042279	1.49	23.03	31	113	Oxidative phosphorylation
41	05216	0.043407	2.18	5.71	10	28	Thyroid cancer
42	00270	0.045874	2.06	6.52	11	32	Cysteine and methionine metabolism
43	04120	0.046424	1.44	26.7	35	131	Ubiquitin mediated proteolysis
44	03030	0.047596	1.96	7.34	12	36	DNA replication