High activation of STAT5A drives peripheral T-cell lymphoma and leukemia

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Supplementary Information

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Supplementary Figure 1. Generation of transgenic mice expressing cS5F under the vav-promoter
A. Schematic representation of the cS5A\textsuperscript{F} (vav-cS5\textsuperscript{F}) construct with restriction sites. B. Western blot on whole cell extracts isolated from Ba/F3 cells starved overnight with or without IL-3 stimulation (5 ng/ml) and Ba/F3 cS5A\textsuperscript{F} cells grown without IL-3 analyzed for FLAG, pYSTAT5 and STAT5A expression. HSC70 served as loading control. Control: Protein from 3T3 cells expressing cS5\textsuperscript{F}-FLAG.
C. cS5A\textsuperscript{F} positive offspring was detected by a 952 bp PCR amplification fragment on genomic DNA
using transgene specific primers. D. Detection of cS5A\textsuperscript{F} copy number in transgenic mice (n=3) by Southern blot signal intensity. 4.7 kB band refers to endogenous Stat5 and a 2.4 kB band refers to the cS5\textsuperscript{F} transgene. DNA from wt littermates (n=2) and from mice expressing cS5\textsuperscript{F} under endogenous promoter were used as controls. E. Western blot on cell lysates of lymph nodes, thymi, gut, liver and kidney from 6-week-old wt, cS5A\textsuperscript{lo} and cS5A\textsuperscript{hi} mice (n=2/genotype) using antibodies to FLAG, phosphotyrosine (Y694)-STAT5 (pYSTAT5) and STAT5A. HSC70 was used as loading control and cS5\textsuperscript{F}-FLAG expressing Ba/F3 cells served as positive control. Representative blots of 4 experiments. F. qPCR analysis showing fold change in mRNA expression of FLAG-tagged STAT5A isolated from FACS sorted CD8\textsuperscript{+}, CD4\textsuperscript{+}, CD19\textsuperscript{+} (spleen), and CD11b\textsuperscript{+} cells (bone marrow) from 8-week-old cS5A\textsuperscript{lo}, cS5A\textsuperscript{hi} and wt mice (n=3/genotype) as well as cultured and FACS-sorted natural killer (NK) cells from 7-week-old wt (n=4) and cS5A\textsuperscript{hi} mice (n=3), mRNA isolated from diseased cS5A\textsuperscript{hi} spleen was used as positive control. Gene expression was calculated relative to the housekeeping gene Gapdh and normalized to the expression of wt. Broken line at 1-fold indicates zero FLAG-tagged STAT5A mRNA expression as in wt cells. G. Left: Hematocrit in peripheral blood obtained in six-week-intervals from wt (n≥6), cS5A\textsuperscript{lo} (n≥8) and cS5A\textsuperscript{hi} (n≥10) mice for 66 weeks (cS5A\textsuperscript{hi} until 42 weeks). Right: flow cytometric analysis of the same peripheral blood samples showing %CD8:%CD4 T-cell ratio (unpaired t-test wt vs cS5A\textsuperscript{lo} P=0.0068, wt vs cS5A\textsuperscript{hi} P=0.0008). H. White blood cell (WBC) count and CD8\textsuperscript{+}:CD4\textsuperscript{+} T-cell ratio in peripheral blood of diseased cS5A\textsuperscript{hi} (n≥10) and hSTAT5B\textsuperscript{N642H} (n≥5) mice and age-matched controls (wt [n≥22], cS5A\textsuperscript{lo} [n≥8], hSTAT5B [n≥12], one-way ANOVA with Tukey’s multiple comparison test compared to wt).
Supplementary Figure 2: High STAT5A activation leads to a neoplastic expansion of mature CD8+ T cells

A. Flow cytometric analysis on relative CD8+ T-cells (P=0.0001), CD4+ T-cells (P<0.0001, both unpaired t-test with Welch’s correction), B-cells (P=0.0001), CD11b+Gr1hi (P=0.0001, unpaired t-test) and NK-cells (P=0.0218, both unpaired t-test with Welch’s correction). B. Relative fraction of (top) CD8+ and CD4+ T-cells, (bottom) CD19+ B-cells and CD11b+Gr1hi granulocytes in peripheral blood, lymph nodes, and bone marrow of diseased cSSAhi mice and wt littermates determined by flow cytometry. All unpaired t-test with Welch’s correction besides CD19+ bone marrow and CD11b+Gr1hi bone marrow unpaired t-test, all P<0.0001, except blood CD11b+Gr1hi cells P=0.0002, bone marrow CD4+ T-cells and CD11b+Gr1hi cells not significant (n.s.), bone marrow B cells P=0.0003). CD11b+Gr1hi cells in lymph nodes were not significantly detectable. C. (Left) Representative dot plots of CD8+ and CD4+ T-cells with gating (shown lymph nodes), (middle) histogram of CD19 expression in wt and cSSAhi splenocytes and (right) dot plots of CD11b+Gr1hi cells and NK-cells with gating (shown spleen, right panel). D. Immunoblot on whole cell lysate of CD8+, CD4+ and CD19+ MACS sorted cells from 10-week-old wt (n=5) and cSSAhi mice (n=4) for FLAG, pYSTAT5 and STAT5 expression. HSC70 served as loading control, cSSAF transfect Ba/F3 cells served as positive and Stat5−/− mouse embryonic fibroblasts (MEFs) as negative control. Representative blot of four experiments. E. Analysis of 100 weeks old cSSAlo and wt mice. lymph nodes weight/body weight and spleen/body weight ratio, F. macroscopic appearance of lymph nodes and spleen (scale bar for 1 cm),
and splenocyte numbers in 100 weeks old wt (n≥2) and cS5Alo (n≥7) mice (lower panel, P=0.04). G. White blood cell (WBC) count in peripheral blood (P=0.0288). H. Statistical summary of flow cytometry staining for CD8+ T-cells on secondary hematopoietic organs (lymph nodes P=0.0098; spleen P=0.0003) (for E-H: All unpaired t-tests, except splenocyte number, white blood cell and CD8+ splenocytes unpaired t-test with Welch’s correction and %CD8 in bone marrow Mann Whitney test P=0.1091). I. Western blot of spleen cell lysates from 70 weeks old cS5Alo (n=2) and wt mice (n=2) showing FLAG, pYSTAT5 and STAT5 expression. cS5Ahhi spleen lysates were used as positive control and a-TUBULIN served as loading control. Representative blot of four experiments J. Tumor weight (left) and tumor incidence per injection (right) 24 days after 1*10⁵ s.c. (subcutaneous) E.G7 cell injection (wt and cS5Ahhi 3 experiments, cS5Alo one experiment, left: Kruskal-Wallis and Dunn’s Multiple Comparison, right Logistic Regression). K. Histograms of CD2, CD3e and CD5 flow cytometry analysis of wt and cS5Ahhi CD8+ splenocytes (FlowJo v10). L. CD25+, M. CD44+, N. CD44+CD62L+ and O. CD44+CD62L- CD8+ T-cells expression in peripheral blood (left), lymph nodes (middle) and bone marrow (right) of diseased cS5Ahhi mice and wt littermates determined by flow cytometry (L. and M. peripheral blood and lymph nodes P<0.0001, bone marrow CD8+CD25+ P<0.0001, CD8+CD44+ P=0.0009, N. peripheral blood P<0.0001, lymph nodes P<0.0001, bone marrow n.s., O. peripheral blood P=0.0005, lymph nodes P<0.0001, bone marrow n.s.). L-O all unpaired t-test with Welch’s correction, L. peripheral blood and lymph nodes unpaired t-test, M. peripheral blood and bone marrow Mann-Whitney test. N (bottom) Representative CD62L/CD44 gating on CD8+ splenocytes by FlowJo. P. Gene set enrichment analysis (GSEA) of genes up- or downregulated in cS5Ahhi compared to cS5Alo CD8+ T-cells (n=5/genotype) on published naïve, memory and effector CD8+ T-cell gene sets. Q. CCR7 expression on CD8+ lymph nodes (top) or spleen (bottom) cells in diseased cS5Ahhi and wt littermates determined by flow cytometry with representative histograms (P=0.0002, unpaired t-test). R. CD62L+CD27CCR7+ and CD62L-CD27CCR7+ CD8+ T-cells in lymph nodes (top, P=0.001 or P<0.0001) and spleen (bottom, P=0.13 or P<0.0001, unpaired t-test). S. Flow cytometric quantification of splenic Treg cells in wt and cS5Ahhi mice with gating strategy (35 weeks P=0.0032, unpaired t-test) and T: thymic γδ T cells (P<0.0001, unpaired t-test).
Supplementary Figure 3. Infiltrating T-cells disrupt lymph nodes and splenic architecture

A. Representative examples of wt lymph nodes and cS5A<sup>hi</sup> lymphomas stained by HE (hematoxylin and eosin, corresponding CD3 staining in Figure 4A). HE (left), CD3 (middle) and Ki67 (right) staining on consecutive cuts of B. skin and D. kidney sections of age-matched wt and diseased cS5A<sup>hi</sup> mice. Scale bars indicate 50, 100 or 200 µm. C. Diseased cS5A<sup>hi</sup> mice with skin lesions. E. Quantification of infiltrating CD3<sup>+</sup> and Ki67<sup>+</sup> cells in skin, lung, liver and kidney in cS5A<sup>hi</sup> and corresponding areas in wt mice (P<0.0001, except for skin %Ki67 positive cells P=0.0078, wt n≥3, cS5A<sup>hi</sup> n≥5, all unpaired t-test).
Supplementary Figure 4. Transplantable CD8⁺ T-cell disease

A. White blood cell (WBC) count in recipient’s blood measured post injection in 4-week-intervals for 16 weeks (n=6/organ source, control n=2, wt lymph nodes n=1, Two Way-ANOVA with Bonferroni Post Test). B. Endpoint analysis (n≥5/group): Spleen/body weight ratio of wt CD8⁺ T-cell recipients (control) and cS5Ahi spleen or lymph nodes CD8⁺ T-cells recipients. C. Percentage of Ly5.2⁺ cells gated on CD8⁺CD3e⁺ T-cells in spleen (left), lymph nodes (middle) and bone marrow (right) of control (wt lymph nodes and wt spleen recipients), spleen- and lymph nodes-derived cS5Ahi CD8 T-cell recipients. 1Way-ANOVA with Tukey’s Multiple Comparison Test (B-C). D. Histological analysis (hematoxylin and eosin (HE) and anti-CD3 staining) on representative areas of spleen (top), liver, kidney and pancreas.
liver (left), kidney (middle) and pancreas (right) from wt or cS5A^{hi} CD8^{+} T-cell (isolated from lymph nodes or spleen) recipients. Scale bars indicate 50, 100 or 200 µm.
Supplementary Figure 5. Expression signature in cS5A<sup>hi</sup> mice

A. RNA-seq heatmap of genes deregulated in comparison of wt, cS5A<sup>lo</sup> and cS5A<sup>hi</sup> CD8<sup>+</sup> T-cells. B. qRT-PCR validation of STAT5 T-cell target genes on mRNA isolated from lymph nodes of diseased cS5A<sup>hi</sup> and age-matched wt mice (Bcl2 P=0.13, Cish P=0.011, Pim1 P=0.01, all unpaired t-test with Welch’s correction, Bcl6 P=0.015, unpaired t-test). C. Summary of gene set enrichment analysis (GSEA) of hallmark gene sets enriched in cS5A<sup>hi</sup> vs wt CD8<sup>+</sup> T-cells. D. Principal component analysis of RNA-seq data of all five genotypes analyzed, wt (n=10), hSTAT5B (n=4), hSTAT5B<sup>N642H</sup>, cS5A<sup>lo</sup> and cS5A<sup>hi</sup> (all n=5). E. Summary of GSEA of T-cell leukemia/lymphoma gene sets positively or
negatively enriched in wt vs cS5A\textsuperscript{hi} CD8\textsuperscript{+} T-cells (false discovery rate [FDR] \( \leq 0.25 \), adj. p-value \( \leq 0.05 \)). F. Summary of Enrichr pathways (KEGG 2016) of genes commonly upregulated in cS5A\textsuperscript{hi} and hSTAT5B\textsuperscript{N642H} or exclusively in hSTAT5B\textsuperscript{N642H} CD8\textsuperscript{+} T-cells, the list of genes can be found in Supplementary Table S5 (only adj. \( P \)-value \( \leq 0.05 \), combined score is \( \log(P\text{-value}) \times Z\text{-score} \)).
Supplementary Figure 6. Expression of STAT5 in PTCL cases

A. STAT5A (top) and STAT5B (bottom) staining of representative PTCL, NOS; AITL; and Cutaneous T-Cell Lymphoma (CTCL) cases with nuclear STAT5A/B staining intensity as summarized in B. Scale bars, 100 or 50 µm. B. Scoring summary of nuclear STAT5A (left) and
STAT5B (right) staining intensity ranging from 1 (low) to 4 (high) in AITL \((n=5)\) and CTCL, \((n=29)\) comprising Mycosis Fungoides (MF), Sézary Syndrome and Lymphomatoid Papulosis). C. Histograms from image analysis based quantification of nuclear STAT5A (left) and nuclear STAT5B (right) staining intensity in the representative images from Figure 8A. Mean intensity (MI) scoring zones are depicted. D. pYSTAT5 staining of PTCL, NOS and AITL case with nuclear STAT5A/B staining intensity 4 and healthy lymph node (hLN) control. E. Positive correlation (right) of STAT5A and STAT5B mRNA expression in PTCL, NOS cases (Pearson correlation \(r=0.92, \ P<0.0001\)). F. STAT5A (left) and STAT5B (middle) mRNA levels of non-diseased human lymph nodes (hLN, \(n=4\)) versus AITL lymphoma tissue \((n=7, \ \text{STAT5A} \ P=0.10, \ \text{STAT5B} \ P=0.22, \ \text{unpaired t-test}\). Mean STAT5A or STAT5B expression in hLN was normalized to 1. Positive correlation (right) of STAT5A and STAT5B mRNA expression in AITL cases (Pearson correlation \(r=0.95, \ P=0.0008\)). G. Statistical summary showing cytoplasmic STAT5A (left) and STAT5B (right) staining intensity, classified as weak positive, positive and strong positive) of 35 PTCL, NOS, 14 AITL, 7 CTCL, 6 MF, and 5 control samples spotted on a tissue microarray.
Supplementary Figure 7. Blockage of JAK/STAT signaling

A. Lymph node cells of 35-week-old wt and cS5A<sup>hi</sup> mice were stimulated with IL-2 (100 U/ml, \(P=0.0011\)), IL-4 (100 ng/ml, \(P \leq 0.0001\)), IL-7 (10 ng/ml, \(P \leq 0.0001\)) or no cytokine (n.s.) for 48 h and \(^{3}H\)thymidine incorporation was measured (\(n=3/\text{genotype in triplicates, all unpaired t-test}\)). B. Western blot for pYSTAT5 and STAT5 on lysates of IL-2-cultured cS5A<sup>hi</sup> lymph node cells sampled at specific time points after IL-2 withdrawal. HSC70 was used as loading control. C. (Top) Viability assay for cS5A<sup>hi</sup> and wt CD8<sup>+</sup> T-cells (\(n=3/\text{genotype}\)) treated in triplicates with Ruxolitinib, Tofacitinib, AC-3-19, 1 µM Bortezomib (positive control set to 0 % cell viability) or DMSO (negative control) in the presence of IL-2 for 72 h. Cell viability was analyzed using CellTiter Glo Assay.
IC₅₀ values (μM) were determined using GraphPad Prism 5 software (GraphPad Software, Inc.), (bottom) numerical summary of the mean IC₅₀ values. D. Human PTCL cell lines (Alk⁻ lines Fe-PD, Mac1 and Mac2A, Alk⁺ lines SU-DHL-1 and SR-786) treated with AC-3-19 (left) or Ruxolitinib (right) for 72 h (n≥3, neg. Control DMSO, 1 μM Bortezomib - positive control set to 0 % cell viability). The KRAS-transformed lung carcinoma cell line A549 cells and Stat5⁻ mouse embryonic fibroblasts (MEFs) were used as control cell lines. Cell viability was measured by CellTiter Glo Assay and IC₅₀ values (μM) were determined using GraphPad Prism 5 software (GraphPad Software, Inc.). E. Western blot on spleen lysates of vehicle or Ruxolitinib treated cS5A<sup>hi</sup> mice for pYSTAT5 and STAT5, HSC70 was used as loading control and diseased cS5A<sup>hi</sup> spleen lysate was used as positive control. F. HE staining on lung sections of vehicle or Ruxolitinib treated cS5A<sup>hi</sup> mice, scale bars represent 400 or 100 μm. G. Histological analysis (Hematoxylin and Eosin (HE) and anti-CD3 staining) on representative liver areas of vehicle or Ruxolitinib treated cS5A<sup>hi</sup> mice. Scale bars represent 400 or 100 μm, respectively.
Supplementary Methods

Generation of cS5A^F transgenic mice
We used the vav-hCD4 hematopoietic vector (HS21/45) and removed the hCD4 insert with restriction endonucleases SfiI and NotI. The hCD4 cassette was replaced by cS5^F-FLAG cDNA taken from pMSCV-cS5^F-FLAG-hCD2. For this, the cS5^F cDNA was excised with EcoRI and subcloned into the plasmid pBluescript II KS (-) (restriction enzymes were purchased from eBioscience, Santa Clara, California, USA, or Fermentas, Waltham, Massachusetts, USA). This plasmid contained an EcoRI site followed by a NotI site in the multiple cloning site. cS5^F was ligated into EcoRI and the NotI site was located at the 3’ end of the insert. The SfiI site was introduced by PCR. The final vav-cS5^F-construct (cS5A^F) was confirmed by restriction digestion (EcoRI, KpnI, BsrHI and HindIII) and sequencing of the cS5^F insert. All primers used are listed in Supplementary Table S1. The cS5A^F construct was linearized with HindIII restriction digestion and gel-purified for pronuclear injection in C57BL/6N zygotes. Embryos were implanted into pseudo-pregnant females. A founder animal was identified by genotyping PCR and Southern blot analysis using DNA isolated from tail biopsies. Genomic PCR was performed with AmpliTaq® DNA Polymerase (Applied Biosystems, Foster City, California, USA) on a Mastercycler (Eppendorf, Hamburg, Germany) using transgene specific primers generating a 952 bp PCR amplification fragment in transgenics and no fragment in wt mice. For Southern blotting, genomic DNA was digested with EcoRI hybridizing with exon 14 of endogenous Stat5a as DNA probe. Transgene presence led to two fragments of 4.7 and 2.4 kb size (wt only 4.7 kb fragment). Two founder lines transmitted to the germline establishing the cS5A^b [B6N-Tg(Vav-cS5F)564Biat] and cS5A^hi [B6N-Tg(Vav-cS5F)565Biat] cohorts. Southern blotting of offspring was used to compare signal intensities of transgene fragments. Mice were viable, fertile and kept on C57BL/6N background.

Testing cS5A^F in vitro
The IL-3 dependent murine B-cell progenitor cell line Ba/F3 was used for testing the cS5A^F construct in vitro. For electroporation, Ba/F3 cells were re-suspended in a density of 5x10^6 cells/800 μl RPMI-1640 without supplements. Electroporation cuvettes with HindIII linearized cS5A^F plasmid DNA and cells were incubated for 5 min at room temperature, put on ice and electroporated at 300 V and 960 μF on a Gene Pulser Xcell™ (Bio-Rad, Hercules, California, USA) and grown in complete RPMI-1640 medium with or without IL-3 (2 ng/ml, ImmunoTools, Friesoythe, Germany) for transgene expression analysis.

Cell Culture
Cells were grown at 37°C and 5% CO2 in standard medium (RPMI1640 or DMEM, details below). Murine cell lines: Ba/F3 cells and its derivatives expressing cS5A^F or BCR-ABL p210^ were cultured in RPMI-1640 medium supplemented with 10% FCS, L-Glutamine (2 mM), penicillin/streptomycin (10 U/ml) (all Gibco, ThermoFisher, Waltham, Massachusetts, USA) with or without IL-3 (2 ng/ml,
ImmunoTools, Friesoythe, Germany). E.G7-OVA and MC-38 cells were kept in RPMI-1640 medium with supplements as described for Ba/F3 cells and 0.05 mM β-Mercaptoethanol (Gibco). Stat5−/− MEFs were prepared from day E13.5-14.5 embryos from STAT5ab+/− mice by mincing the carcass after removal of head and internal organs, digesting in 0.05% trypsin/0.53 mM EDTA (Gibco) for 10 min at 37 °C, re-suspending in complete growth medium (high glucose DMEM with 10% FCS, 2 mM L-Glutamine, 100 U/ml Penicillin-Streptomycin, 10 μg/ml Gentamicin, 0.1 M MEM Non-Essential Amino Acids, 55 μM β-Mercaptoethanol, all Gibco) and grown until confluent. The cS5Ah CTL lines were established from lymph node single cells suspensions from diseased cS5Ah mice in presence of 1 μg/ml anti-mouse CD3e (BD Pharmingen, Purified NA/LE hamster anti-mouse, Franklin Lakes, New Jersey, USA) and 100 U/ml IL-2 (ImmunoTools), corresponding wt T-cells were kept in culture for 2 weeks. Murine CTLs were grown in complete RPMI-1640 medium (10% FCS, 2 mM L-Glutamine, 10 U/ml Penicillin/Streptomycin; all Gibo) containing in addition 10 mM HEPES, 1xMEM Non-Essential Amino Acids, 50 μM β-Mercaptoethanol (all Gibco), 1 mM Sodium Pyruvate (Sigma-Aldrich, St. Louis, Missouri, USA), and 100 U/ml IL-2 (ImmunoTools). Medium was exchanged twice/week and CD8 and CD3e surface marker expression was checked regularly using flow cytometry.

Human cell lines: SR-786 (ACC-369) and SU-DHL-1 (ACC-356; both ALK+ ALCL) were from DSMZ (Braunschweig, Germany). Fe-PD (ALK ALCL), Mac1 and Mac2A were obtained from O.M. The lung carcinoma cell line A549 was supplied from ATCC. Mac1/2A cells have been derived from the same donor patient, Mac1 were established in the indolent cutaneous lymphoma phase, while Mac2A were established in advanced disease stage from cutaneous tumor nodules with anaplastic morphology (ALK ALCL).5,6 Human cell lines were grown in complete RPMI-1640 medium with 10 mM HEPES (Gibco).

**mRNA isolation and qRT-PCR**

For murine cells, mRNA was isolated from cell pellets using TRIzol® (Invitrogen, ThermoFisher, Waltham, Massachusetts, USA). For patient derived samples, deparaffinization of formalin-fixed, paraffin embedded (FFPE) sections was performed using xylol extraction (Sigma) followed by isolation of RNA using the miRNeasy kit (Qiagen, Hilden, Germany) according to manufacturer’s protocol. First strand cDNA synthesis from isolated RNA was done using the RevertAid™ H Minus First Strand cDNA Synthesis Kit (Fermentas) using gene specific primers for FFPE samples (see supplementary Table S1). qRT-PCR was performed on a RealPexcycler (Eppendorf) using SYBR green (Roche, Basel, Switzerland) incorporation and mRNA levels were normalized for murine or human Glyceraldehyde 3-phosphate dehydrogenase (Gapdh), respectively. PTCL FFPE samples with GAPDH ΔCt values >30 were excluded (RNA degradation). Relative fold expression was calculated using the ΔCt method. Primer pairs are listed in Supplementary Table S1.
**Immunohistochemistry**

Mouse organs were fixed overnight in 4% phosphate buffered formaldehyde solution (Roti® Histofix, Carl Roth, Karlsruhe, Germany), dehydrated, paraffin embedded and cut. For immunohistochemical stainings, heat-mediated antigen retrieval was performed in citrate buffer at pH 6.0 (S1699; Dako, Agilent Technologies, Santa Clara, California, USA) and stained with antibodies against CD3, and Ki67 (listed in Supplementary Table S4) using standard protocols.

Human lymphoma cases were incubated overnight at 4 °C with STAT5A, STAT5B, or pYSTAT5 antibodies (listed in Supplementary Table S4). For STAT5A and STAT5B, a polymer-based detection system was used (Lab Vision™ UltraVision™ LP Detection System, Thermo Scientific), for pYSTAT5 an Avidin-Biotin system was used. The specific signals were amplified with AEC under visual control followed by counterstaining with Hemalaun.

For Ki67 and CD3 quantification, at least five different high-power field sections (x200 objective) of infiltrated organ areas or corresponding wildtype (wt) areas per mouse and genotype were compared using HistooQuest (TissueGnostics GmbH, Vienna, Austria) quantification software as described. For nuclear STAT5A/B staining intensity quantification, high-power field sections (x200 objective) were obtained for each sample and analyzed with HistooQuest or StrataQuest (TissueGnostics GmbH) quantification software as described. In brief, cut-offs (to differentiate between positive and negative cells) were set for all samples and the resulting positive nuclear STAT5A/B intensities were normalized to Hemalaun staining intensity and used for scoring based on the following intervals: STAT5A score 1 MI≤55, score 2 55<MI≤75, sore 3 75<MI≤95, score 4 95<MI≤115; STAT5B score 1 MI≤45, score 2 45<MI≤80, sore 3 80<MI≤115, score 4 115<MI≤150 or scoring weak positive, positive, strong positive.

**RNA-seq processing, analysis and gene set enrichment analysis**

CD8⁺ T-cells were enriched using CD8⁺ Magnisort kit (eBioscience, Santa Clara, California, USA) and mRNA was isolated using Trizol (Sigma-Aldrich, St. Louis, Missouri, USA) in combination with RNeasy mini kit (Qiagen, Hilden, Germany). mRNA library preparation (SENSE mRNA-Seq Library preparation) and RNA sequencing was performed with Illumina HiSeq-2500 at VBCF NGS Unit (www.vbcf.ac.at). Adapter trimming and removal of low quality bases was performed using cutadapt. After alignment of reads against contaminating sequences (mitochondrial and ribosomal DNA) remaining reads were aligned against GRCm37 using transcriptome guided alignment with TopHat version 1.4.1. Following, htseq-count with mode union was used to get gene counts for union gene models. Then differentially expressed genes (log2 fold change > 2 and FDR q-adjust < 0.1) were determined using DESeq2 version 1.12.4.

For heatmaps, centered and scaled rlog transformed library size normalized counts were visualized using the heatmap.2 function of R package gplots version 3.0.1.

For pathway analysis, the Enrichr web tool was applied. The command-line version of GSEA was used for gene-set enrichment analysis (GSEA), and log-2 fold changes between control and experimental condition were used as ranking metric.
Gene sets were downloaded from the Molecular Signature Database v5.0 (http://www.broadinstitute.org/gsea/msigdb/index.jsp) or compiled from literature.\textsuperscript{11,12}

**Hematocytometry and flow cytometry**  
Blood was obtained by tail incisions or *vena facialis* puncture collected in EDTA-tubes (Mini-Collect K3EDTA tubes, Greiner Bio-One, Kremsmuenster, Austria) and smears were stained using Modified Wright Staining. White blood cell count and hematocrit were measured using an animal blood counter (sciI Vet abc, Viernheim, Germany). For flow cytometry, erythrocytes were lysed using Gay's solution (10 mM KHCO\textsubscript{3} and 75 mM NH\textsubscript{4}Cl, pH 7.4). Flow cytometry was used in order to determine cellular components of lymph nodes, thymus, bone marrow and spleen. Single cell suspensions were prepared by mincing organs through a 70 µm-cell strainer (BD Biosciences, Franklin Lakes, USA). All antibodies used in this study are listed in Supplementary Table S3. All analyses were performed on the BD FACS Canto II™ instrument and calculated with FACSDiva software (BD Biosciences) or FlowJo.

**Viability assay**  
5×10\textsuperscript{3} murine or human cells were seeded in triplicates in 96-well plates. In case of murine cells, 100 U/ml IL-2 was present in the medium. The STAT5 inhibitor AC-3-19, Ruxolitinib (Cayman Chemical Company, Ann Arbor, USA), Tofacitinib or 1 µM Bortezomib (both Selleckchem, Houston, USA) was added for 72 h. Bortezomib served as positive control for cell death (well set to 0 % viability). CellTiter-Blue\textsuperscript{®} or CellTiter-Glo\textsuperscript{®} (Promega, Madison, USA) reagent was used to determine viability measured on an EnSpire plate reader (PerkinElmer, Waltham, USA). IC\textsubscript{50} values were determined using GraphPad Prism\textsuperscript{®} 5 (GraphPad Software, San Diego, California) by non-linear regression.

**Western blotting**  
Western blotting was done according to standard methods. Nitrocellulose membranes (Amersham™ Protran, GE Healthcare, Chicago, USA) were incubated with antibodies listed in Supplementary Table S2. HSC70 or β-ACTIN served as loading control.

**MACS and FACS sorting**  
Magnetic cell sorting (MACS) was done on single cell suspensions prepared from lymph nodes or spleens using CD4 (L3T4), CD8a (Ly-2), CD19 or CD49b (DX5) microbeads (all anti-mouse and purchased from Miltenyi Biotec, Bergisch Gladbach, Germany) following instructions. FACS sorting of spleen and bone marrow cells or MACS-sorted, 5 days cultured NK cells was performed at 4°C on a FACSariaII (BD Biosciences) equipped with a 488, 561, 633 and 395 nm laser and FACSDiva software (BD Biosciences) using CD4, CD8, CD19, CD11b, CD3e and NK1.1 antibodies.
Intracellular staining

Intracellular staining of FOXP3 on spleen and lymph nodes was carried out using the T<sub>reg</sub> staining buffer kit (eBioscience Santa Clara, California, USA) and CD4, CD8 and CD25 antibodies (listed in Supplementary Table S3).

E.G7-OVA and MC-38 tumor growth

1x10<sup>6</sup> or 1x10<sup>5</sup> E.G7-OVA or MC-38 in 100 µl PBS were injected s.c. into both flanks of 10-week-old wt, cS5A<sup>lo</sup> and cS5A<sup>hi</sup> mice, which were monitored daily. Tumor weights were determined 12 (for 1x10<sup>6</sup> cells), 24 (for 1x10<sup>5</sup> cells) or 18 days (MC-38) after injection.

<sup>[3]H</sup>-thymidine incorporation assay

<sup>[3]H</sup>-thymidine incorporation assay was done as described previously.<sup>13</sup> In brief, 5x10<sup>4</sup> lymph nodes cells or CD8<sup>+</sup> MACS sorted splenic T-cells isolated from 25-week-old wt and cS5A<sup>hi</sup> mice were plated in triplicates in 96 well plates in the presence of 0.1 µCi/well <sup>[3]H</sup>-thymidine (PerkinElmer, Waltham, Massachusetts, USA) supplemented with IL-2 (100 U/ml, Proleukin<sup>®</sup>, Novartis, Basel, Switzerland), IL-4 (100 ng/ml, 404-ML), IL-7 (10 ng/ml, both R&D Systems, Minneapolis, USA) or without cytokines for 48 h. Incorporated radioactive counts were measured using TopCount NXT Microplate Scintillation and Luminescence Counter (PerkinElmer, Waltham, Massachusetts, USA).

CD8<sup>+</sup> T-cell transfer

CD8<sup>+</sup> T-cells were isolated by positive MACS separation (see section MACS sorting) from single cell suspensions of lymph nodes or spleens. 2x10<sup>6</sup> cells were injected i.v. into Ly5.1<sup>+</sup> mice (two recipients/organ). In four-week-intervals, blood parameters were measured (scil Vet abc, Viernheim, Germany) and flow cytometric staining was performed to follow the donor-derived cell contribution. Diseased mice were euthanized, hematopoietic organs and peripheral blood analyzed using flow cytometry (CD4, CD8, CD3e, Ly5.1, Ly5.2) for the contribution of Ly5.1<sup>+</sup> and Ly5.2<sup>+</sup> cells to T-cells. Histopathology of spleen, lung, liver, kidney and pancreas was performed using HE and CD3 staining.

In vivo Ruxolitinib treatment

25 weeks old cS5A<sup>hi</sup> mice were treated with Ruxolitinib (Chemietek, Indianapolis, Indiana) twice a day at 45 mg/kg dosage via oral gavage for 30 days. Ruxolitinib was dissolved in DMSO (Sigma-Aldrich. St, Louis, Missouri, USA) and subsequently diluted in 0.5% methylcellulose (w/v, Sigma-Aldrich).
Supplementary Tables

Supplementary Table S1: Primers
All primers were obtained from Eurofins MWG/Operon (Ebersberg, Germany). All sequences are written in 5’ to 3’ direction.

| Primers for PCR (cloning) | Sequence                        |
|---------------------------|---------------------------------|
| SfiIStat5a for            | GCAGGCCCCGTACGCGCGAATTCGACTCAACAACCCAG |
| BclIStat5a rev            | GCAAACTGAGCTTGGATCCCG            |
| NolI ER rev               | GCATCAGCGCCGCCGCCCCCGGTCAGATCGGTGG |

Southern blotting primers

| Stat5a-exon 14 for        | GGCAGGGTGCACATTGCTGTC           |
| Stat5a-exon 14 rev        | CCGTTGAACCTGGAACCAGGA           |

Genotyping Primers

| cS5 frw                   | AGGCGACCATCATCAGCGAGCG         |
| cS5 rev                   | GAATGGAGAATAATCGTGCGCG         |

qPCR primers - murine

| Gapdh frw                 | AGAAGGTGGGTAAGCAGGCACT         |
| Gapdh rev                 | CGGCATCGAAGTTGGAAGAGTG         |
| Stat5a frw                | ACGCCGCCCATGCGACGTC            |
| FLAG rev                  | CTTGCATCGTCGTCCTTGATGTC        |
| Stat5a rev                | CGAAGGCAAAACTGCGACGGC          |
| Stat5a rev                | TCTCCGATCTCTGCTGTGATCAG        |
| Bcl2 frw                  | ACTGAATCCTGAGCCCGCATC          |
| Bcl2 rev                  | GGAGAAATCAAAACAGAGGTCG         |
| Bcl6 frw                  | GATACAGCTGTCAGCGGG             |
| Bcl6 rev                  | AGTTTCTAGGAAGAGGCGGGA          |
| Cis frw                   | CTGGGACTCTAACCTGCTGTC          |
| Cis rev                   | TAGGCAACCGAGGTCAC              |
| Pim1 frw                  | TTCTCCACCGCGACATCAA            |
| Pim1 rev                  | TAGGCAATCCACTCTGGAGGAC         |

qPCR primers - human

| STAT5a frw                | TCTCCCTCTGACTTCAAACAGCG        |
| STAT5a rev                | ACCACCTCTAAGCTGGATAGCC         |
| STAT5a_RevTrans1          | AGGGGCGAGAGGCGGGAG             |
| STAT5b frw                | GATCAAGCAAGTGGTCC             |
| STAT5 rev                 | CCAGATCGAAGTCCCCATCGG          |
| STAT5b_RevTrans1          | CCGGCTACGCTCGATCTGTG          |
| GAPDH frw                 | TCTTTTGCGTCCCGAGCGGGA         |
| GAPDH rev                 | GCGCCCAATACGACCAAAATCCGTT     |
| GAPDH_RevTrans1           | TGACCAGCGCCCAATACGAC          |
### Supplementary Table S2: Western Blot Antibodies

| Antibody                                      | Company                                | Catalogue No. | Dilution |
|------------------------------------------------|----------------------------------------|---------------|----------|
| Monoclonal rabbit anti-mouse phospho-Stat5    | Invitrogen (Carlsbad, California, USA) | 716900        | 1:1,000  |
| Monoclonal mouse anti-FLAG                    | Sigma-Aldrich (St. Louis, Missouri, USA) | F3165         | 1:2,000  |
| Polyclonal rabbit anti-Stat5 (N-20)           | Santa Cruz (Dallas, Texas, USA)        | Sc-836        | 1:5,000  |
| Monoclonal Rabbit anti-STAT5A antibody        | Epitomics (Burlingame, California, USA) | 1289-1        | 1:1,000  |
| Polyclonal rabbit anti-mouse Stat5a           | Self-made rabbit serum against the extreme C-terminus |              | 1:5,000  |
| Monoclonal mouse anti-mouse HSC-70            | Santa Cruz (Dallas, Texas, USA)        | sc-7298       | 1:10,000 |
| Purified mouse anti-Stat5 antibody            | Becton Dickinson (Franklin Lakes, New Jersey, USA) | 610191        | 1:2,000  |
| Monoclonal mouse anti-mouse β-actin           | Sigma-Aldrich (St. Louis, Missouri, USA) | A5136         | 1:50,000 |
| α-Tubulin                                     | Santa Cruz (Dallas, Texas, USA)        | Sc-32293      | 1:10,000 |
| ECL™ Anti-mouse IgG, Horseradish Peroxidase linked whole antibody (from sheep) | GE Healthcare (Chicago, Illinois, USA) | NA931V        | 1:10,000 |
| ECL™ Anti-rabbit IgG, Horseradish Peroxidase linked whole antibody (from sheep) | GE Healthcare (Chicago, Illinois, USA) | NA934V        | 1:10,000 |
Supplementary Table S3: Flow Cytometry Antibodies

All antibodies were obtained from eBioscience (Santa Clara, California, USA) unless indicated otherwise.

| Antigen       | Fluorochrome                  | Catalogue No.          |
|---------------|-------------------------------|------------------------|
| CD117 (c-Kit) | PE-Cy5, APC                   | 15-1171-83, 17-1171-82 |
| CD11b         | FITC, eFluor® 450             | 11-0112-82, 48-0112-82 |
| CD16/32 (Fc-block) | Unconjugated, PE   | 14-0161-82, 12-0161-81 |
| CD19          | PE, eFluor® 450               | 12-0193-82, 48-0193-82 |
| CD197 (CCR7)  | eFluor® 450                   | 48-1971-80             |
| CD2           | PE                            | 12-0021-81             |
| CD25          | APC                           | 17-0251-81             |
| CD27          | PE                            | 12-0271-82             |
| CD3           | eFluor® 450                   | 48-0032-82             |
| CD3e          | PerCP Cy5.5                   | 45-0031-82             |
| CD4           | FITC, PE                      | 11-0041-82, 12-0041-82 |
| CD44          | PE                            | 12-0441-82             |
| CD45.1 (Ly5.1)| PE                            | 12-0453-82             |
| CD45.2 (Ly5.2)| APC                          | 17-0454-82             |
| CD45R (B220)  | eFluor® 450, PerCP Cy5.5      | 48-0452-82, 45-0452-82 |
| CD49b         | APC, eFluor® 780              | 17-5971-82, 47-5971-82 |
| CD5           | APC                           | 17-0051-81             |
| CD62L         | APC                           | 17-0621-81             |
| CD8a          | FITC, PE, PerCP Cy5.5, APC    | 11-0081-82, 12-0081-82, 45-0081-82, 17-0081-82 |
| FOXP3         | PE                            | 12-5773-80A            |
| Lineage Panel | Biotinylated, APC-Cy7 or eFluor® 450 | BD 559971 |
| Ly6.G         | APC                           | 17-9668-80             |
| Ly6.G (Gr1)   | APC, eFluor® 450              | 17-5931-82, 48-5931-82 |
| Ly-6A/E (Sca-1)| PE-Cy7, FITC                 | 25-5981-82, 11-5981-81 |
| NK1.1         | PE-Cy8n®7                     | 25-5941-82             |
| Streptavidin  | PerCP Cy5.5, APC-eFluor® 780  | 45-4317-82, 47-4317-82 |
| TER-119       | eFluor 450                    | 48-5921-82             |
| TCRγδ         | FITC                          | 11-5711-81             |

Supplementary Table S4: Immunohistochemistry Antibodies

| Antibody | Company                                  | Catalogue No. | Dilution |
|----------|------------------------------------------|---------------|----------|
| CD3      | ThermoFisher Scientific (Waltham, Massachusetts, USA) | RM-9107-S0   | 1:300    |
| Ki67     | Novocastra, Leica Biosystems (Wetzlar, Germany) | NCL-Ki67p    | 1:1,000  |
| STAT5A   | Epitomics (Burlingame, California, USA)   | 1289-1        | 1:50     |
| STAT5B   | Santa Cruz (Dallas, Texas, USA)           | sc-1656       | 1:200    |
| pYSTAT5  | Cell Signaling Technologies (Danvers, Massachusetts, USA) | 9359         | 1:200    |
## Supplementary Table S5: GSEA for PTCL-NOS with cytotoxic T-cell features (cS5Ai)

| row_0 | GZMB | 1 0.081905 | Yes | row_37 | row_40 | IL10 | 9134 | 0.080061 | No |
| row_1 | KLRC3 | 15 0.140442 | Yes | row_34 | row_41 | IL21R | 9172 | 0.078939 | No |
| row_2 | KLRC2 | 17 0.199820 | Yes | row_33 | row_42 | LY96 | 9197 | 0.078748 | No |
| row_3 | KLRC4 | 18 0.259268 | Yes | row_3 | row_43 | TNFRSF1A | 9437 | 0.063726 | No |
| row_4 | TBX21 | 40 0.299721 | Yes | row_75 | row_44 | CTSA | 9622 | 0.052720 | No |
| row_5 | CCL5 | 46 0.339709 | Yes | row_72 | row_45 | VSIG4 | 9866 | 0.037852 | No |
| row_6 | IL18RAP | 47 0.379923 | Yes | row_70 | row_46 | ILF2 | 10181 | 0.018401 | No |
| row_7 | FASLG | 48 0.419827 | Yes | row_69 | row_47 | CD84 | 10425 | 0.004230 | No |
| row_8 | CXCR3 | 86 0.448979 | Yes | row_68 | row_48 | CD226 | 10728 | -0.013688 | No |
| row_9 | EOMES | 92 0.478837 | Yes | row_67 | row_49 | IDO1 | 10802 | -0.015485 | No |
| row_10 | SLAMF7 | 105 0.505789 | Yes | row_66 | row_50 | FCGR3B | 10882 | -0.017593 | No |
| row_11 | IL2RB | 139 0.528803 | Yes | row_65 | row_51 | CTSK | 11089 | -0.028282 | No |
| row_12 | CX3CR1 | 192 0.546449 | Yes | row_64 | row_52 | KLRC1 | 11431 | -0.047828 | No |
| row_13 | IL10RB | 232 0.563071 | Yes | row_63 | row_53 | ILF3 | 11600 | -0.054976 | No |
| row_14 | TNFSF14 | 295 0.575283 | Yes | row_52 | row_54 | TGFRB1 | 11677 | -0.055570 | No |
| row_15 | IFNGR1 | 325 0.588911 | Yes | row_51 | row_55 | IL27RA | 11727 | -0.054171 | No |
| row_16 | PRF1 | 326 0.604561 | Yes | row_50 | row_56 | CCR1 | 11788 | -0.053439 | No |
| row_17 | GZMA | 512 0.603479 | Yes | row_49 | row_57 | SLAMF8 | 11803 | -0.049460 | No |
| row_18 | TNFSF10 | 573 0.610325 | Yes | row_48 | row_58 | CCL18 | 11835 | -0.046610 | No |
| row_19 | CTSD | 787 0.607475 | No | row_47 | row_59 | CTSS | 12078 | -0.057981 | No |
| row_20 | CTSB | 880 0.606929 | No | row_46 | row_60 | CTSB | 12200 | -0.060584 | No |
| row_21 | CD47 | 1178 0.593344 | No | row_45 | row_61 | CTSC | 12401 | -0.068123 | No |
| row_22 | FADD | 1200 0.598992 | No | row_44 | row_62 | IL1R2 | 12427 | -0.063326 | No |
| row_23 | SH2D1A | 1716 0.568427 | No | row_43 | row_63 | KLRC1 | 12462 | -0.059076 | No |
| row_24 | TGFBR1 | 1920 0.559195 | No | row_42 | row_64 | IL31RA | 12575 | -0.059593 | No |
| row_25 | GZMK | 2056 0.554456 | No | row_41 | row_65 | CCL8 | 12989 | -0.080144 | No |
| row_26 | CD8B | 2226 0.546972 | No | row_40 | row_66 | IL1R1 | 13044 | -0.074904 | No |
| row_27 | KLRK1 | 2266 0.548527 | No | row_39 | row_67 | XCL2 | 13047 | -0.060615 | No |
| row_28 | IFNAR1 | 2505 0.535746 | No | row_38 | row_68 | XCL1 | 13048 | -0.056986 | No |
| row_29 | IL2RG | 4143 0.422963 | No | row_37 | row_69 | CXCL10 | 13205 | -0.057855 | No |
| row_30 | TNFRSF1B | 4180 0.422223 | No | row_36 | row_70 | CXCR4 | 13330 | -0.055689 | No |
| row_31 | CD8A | 4931 0.370750 | No | row_35 | row_71 | CXCL9 | 13363 | -0.046904 | No |
| row_32 | KIR3DL2 | 6549 0.257546 | No | row_34 | row_72 | IL1RN | 13650 | -0.053331 | No |
| row_33 | KIR3DL3 | 6550 0.257546 | No | row_33 | row_73 | TNFRSF12A | 13692 | -0.042076 | No |
| row_34 | KIR2DL1 | 6551 0.257546 | No | row_32 | row_74 | CD244 | 13704 | -0.028570 | No |
| row_35 | KIR2DL3 | 6552 0.257546 | No | row_31 | row_75 | SLAMF6 | 13833 | -0.021426 | No |
| row_36 | KIR3DL1 | 6553 0.257546 | No | row_30 | row_76 | IL18 | 13950 | -0.011375 | No |
| row_37 | KIR2DL4 | 6554 0.257546 | No | row_29 | row_77 | IL13RA1 | 14005 | 0.004201 | No |
| row_38 | CXCL11 | 7701 0.177693 | No | row_28 | row_78 | IFNGR2 | 14055 | 0.021493 | No |
| row_39 | CCL2 | 8835 0.099500 | No | row_27 | row_79 | No | 0.177693 | No | 0.021493 | No |

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Supplementary Table S6: Enrichr

| Term                                                                 | Overlap | P-value   | Adj. P-value | Z-score   | Combined Score | Genes                                      |
|----------------------------------------------------------------------|---------|-----------|--------------|-----------|----------------|--------------------------------------------|
| **upregulated exclusively in hSTAT5B\textsuperscript{N642H}**        |         |           |              |           |                | SMAD3, CDKN2C, CDKN2A, PLK1, BUB1B,        |
| Cell cycle_Homo sapiens_hsa04110                                     | 17 of 124 | 1.817E-11 | 2.653E-09    | -1.73320  | 42,864         | CDC25C, GADD45G, CDC20, CCNA2, CCNB2,      |
| p53 signaling pathway_Homo sapiens_hsa04115                          | 9 of 69  | 1.703E-06 | 0.00012      | -1.73739  | 23,078         | CCNB2, CCNB1, RRM2, CDKN2A, ZMAT3, CHEK1,|
| Pyrimidine metabolism_Homo sapiens_hsa00240                          | 8 of 105 | 0.00032   | 0.01544      | -1.73479  | 13,975         | CDK1, BAX, GADD45G                        |
| Oocyte meiosis_Homo sapiens_hsa04114                                 | 8 of 123 | 0.00091   | 0.03327      | -1.80427  | 12,631         | CDC20, CCNB2, CCNB1, SGOL1, ESPL1, PLK1,  |
| Viral carcinogenesis_Homo sapiens_hsa05203                           | 10 of 205| 0.00199   | 0.04845      | -1.85515  | 11,537         | CDC20, CCNA2, CDKN2A, CHEK1, CDK1, BAX,   |
| **upregulated in c55A\textsuperscript{hi} and hSTAT5B\textsuperscript{N642H}** |         |           |              |           |                | HIST1H2BE, HIST1H4D, CCR4, CCR3          |
| Cytokine-cytokine receptor interaction_Homo sapiens_hsa04060         | 15 of 265 | 1.045E-07 | 1.411E-05    | -1.87718  | 30,174         | CX3CR1, CSF2, IL10RA, IL18RAP, IFNG, CXCR3,|
| Jak-STAT signaling pathway_Homo sapiens_hsa04630                     | 9 of 158 | 3.767E-05 | 0.00254      | -1.90533  | 19,409         | IL2RA, CCL5, IL2RB, TNFSF8, CCR5, IL18R1,|
| Inflammatory bowel disease (IBD)_Homo sapiens_hsa05321                | 5 of 65  | 0.00054   | 0.02432      | -1.89732  | 14,274         | IL18RAP, IFNG, TBX21, IL18R1, IL12RB2     |
## Supplementary Table S7: Reported mutations in human T-cell lymphoma cell lines

| Cell Line | Type                                                                 | JAK/STAT mutations          | Other relevant mutations                  |
|-----------|----------------------------------------------------------------------|------------------------------|-------------------------------------------|
| SR786     | ALCL cell line of T-cell type derived from pediatric CD30+ lymphoma  |                              | NPM1-ALK, missense TP53                   |
| Mac1      | ALCL, Mac2A sister cell line, at indolent stage - different immunological, functional features | PCM1-JAK2, JAK3 V722I       |                                           |
| Mac2A     | ALCL cell line (of T-cell type) derived from aggressive ALCL post-Hodgkin lymphoma and post-CTCL, representing advanced skin CD30+ Mycosis Fungoides | PCM1-JAK2, JAK3 V722I, SOCS1 G78R/D105N |                                           |
| Su-DHL-1  | ALCL cell line derived from child with malignant histiocytosis      |                              | NPM1-ALK, missense TP53 and DNMT3A mutation |
| Fe-PD     | ALCL cell line                                                        | STAT3 G618R, JAK1 G1097V    |                                           |
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