abcam

Product datasheet

Human IKZF1 knockout Jurkat cell line ab274920

3 Images

Overview

Product name	Human IKZF1 knockout Jurkat cell line
Parental Cell Line	Jurkat
Organism	Human
Mutation description	Knockout achieved by CRISPR/Cas9; X = 2 bp insertion, 1 bp insertion; Frameshift: 100%
Passage number	<20
Knockout validation	Next Generation Sequencing (NGS), Western Blot (WB)
Tested applications	**Suitable for:** WB, Next Generation Sequencing
Biosafety level	1
General notes	**Recommended control:** Human wild-type Jurkat cell line (**ab271143**). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1. **Cryopreservation cell medium:** Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose. **Culture medium:** RPMI + 10% FBS **Initial handling guidelines:** Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability. 1. Thaw the vial in 37°C water for bath approximately 1-2 minutes. 2. Transfer the cell suspension (0.8 mL) to a 15 mL/50 mL conical sterile polypropylene centrifuge tube containing 8.4 mL pre-warmed culture medium, wash vial with an additional 0.8 mL culture medium (total volume 10 mL) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 mL represents minimum recommended dilution. 20 mL represents maximum recommended dilution. 3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of $1x10^5$ cells/mL. Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37°C incubator with 5% CO_2. Cultures should be monitored daily. **Subculture guidelines:** All seeding densities should be based on cell counts gained by established methods. A guide seeding density of $1x10^5$ cells/mL is recommended. Do not allow the cell density to exceed $3x10^6$. This product is subject to limited use licenses from The Broad Institute, ERS Genomics Limited

and Sigma-Aldrich Co. LLC, and is developed with patented technology. For full details of the licenses and patents please refer to our **limited use license** and **patent pages**.

We will provide viable cells that proliferate on revival.

Properties

Number of cells	1 x 10^6 cells/vial, 1 mL
Adherent /Suspension	Suspension
Tissue	Blood
Cell type	T cell lymphoblast-like
Disease	Non-Hodgkin Lymphoma
Gender	Male
Mycoplasma free	Yes
Storage instructions	Shipped on Dry Ice. Store in liquid nitrogen.
Storage buffer	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

Target

Function	Transcription regulator of hematopoietic cell differentiation (PubMed:17934067). Binds gamma-satellite DNA (PubMed:17135265, PubMed:19141594). Plays a role in the development of lymphocytes, B- and T-cells. Binds and activates the enhancer (delta-A element) of the CD3-delta gene. Repressor of the TDT (fikzfterminal deoxynucleotidyltransferase) gene during thymocyte differentiation. Regulates transcription through association with both HDAC-dependent and HDAC-independent complexes. Targets the 2 chromatin-remodeling complexes, NuRD and BAF (SWI/SNF), in a single complex (PYR complex), to the beta-globin locus in adult erythrocytes. Increases normal apoptosis in adult erythroid cells. Confers early temporal competence to retinal progenitor cells (RPCs) (By similarity). Function is isoform-specific and is modulated by dominant-negative inactive isoforms (PubMed:17135265, PubMed:17934067).
Tissue specificity	Abundantly expressed in thymus, spleen and peripheral blood Leukocytes and lymph nodes. Lower expression in bone marrow and small intestine.
Involvement in disease	Defects in IKZF1 are frequent occurrences (28.6%) in acute lymphoblasic leukemia (ALL). Such alterations or deletions lead to poor prognosis for ALL. Chromosomal aberrations involving IKZF1 are a cause of B-cell non-Hodgkin lymphomas (B-cell NHL). Translocation t(3;7)(q27;p12), with BCL6.
Sequence similarities	Belongs to the Ikaros C2H2-type zinc-finger protein family. Contains 6 C2H2-type zinc fingers.
Domain	The N-terminal zinc-fingers 2 and 3 are required for DNA binding as well as for targeting IKFZ1 to pericentromeric heterochromatin. The C-terminal zinc-finger domain is required for dimerization.
Post-translational modifications	Phosphorylation controls cell-cycle progression from late G(1) stage to S stage. Hyperphosphorylated during G2/M phase. Dephosphorylated state during late G(1) phase. Phosphorylation on Thr-140 is required for DNA and pericentromeric location during mitosis. CK2 is the main kinase, in vitro. GSK3 and CDK may also contribute to phosphorylation of the C-terminal serine and threonine residues. Phosphorylation on these C-terminal residues reduces the

	DNA-binding ability. Phosphorylation/dephosphorylation events on Ser-13 and Ser-295 regulate TDT expression during thymocyte differentiation. Dephosphorylation by protein phosphatase 1 regulates stability and pericentromeric heterochromatin location. Phosphorylated in both lymphoid and non-lymphoid tissues (By similarity). Phosphorylation at Ser-361 and Ser-364 downstream of SYK induces nuclear translocation. Sumoylated. Simulataneous sumoylation on the 2 sites results in a loss of both HDAC-dependent and HDAC-independent repression. Has no effect on pericentromeric heterochromatin location. Desumoylated by SENP1. Polyubiquitinated.
Cellular localization	Cytoplasm; Nucleus. In resting lymphocytes, distributed diffusely throughout the nucleus. Localizes to pericentromeric heterochromatin in proliferating cells. This localization requires DNA binding which is regulated by phosphorylation / dephosphorylation events and Nucleus. In resting lymphocytes, distributed diffusely throughout the nucleus. Localizes to pericentromeric heterochromatin in proliferating cells. This localization requires DNA binding which is regulated by phosphorylation / dephosphorylation events (By similarity).
Form	There are 7 isoforms produced by alternative splicing.

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab274920 in the following tested applications.

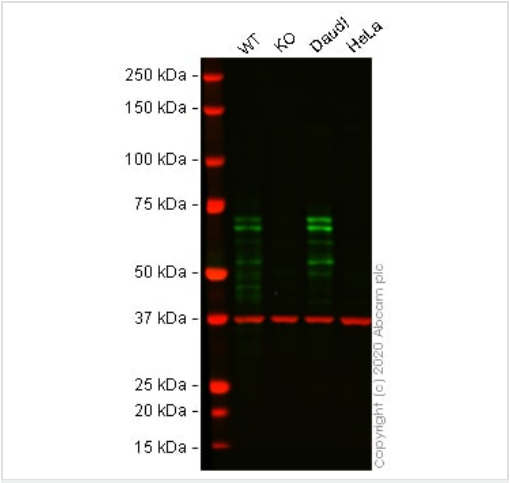
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration.
Next Generation Sequencing		Use at an assay dependent concentration.

Images

Next Generation Sequencing - Human IKZF1 knockout Jurkat cell line (ab274920)

2 bp deletion (allele 1) and 1 bp deletion (allele 2) after Arg50 of the WT protein



Western blot - Human IKZF1 knockout Jurkat cell line (ab274920)

All lanes : Anti-Ikaros antibody [EPR13790] (**ab191394**) at 1/10000 dilution

Lane 1 : Wild-type Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate
Lane 2 : IKZF1 knockout Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate
Lane 3 : Daudi (Human Burkitt's lymphoma cell line) whole cell lysate
Lane 4 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

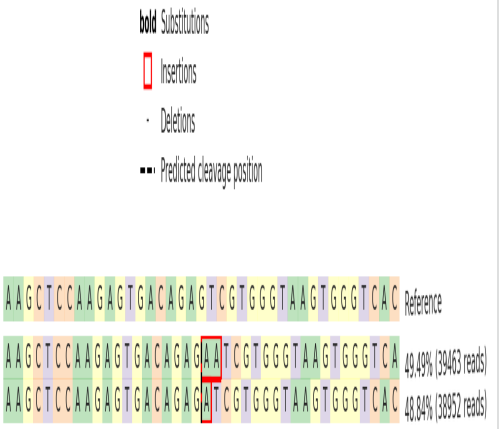
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 50-70 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab191394** observed at 50-70 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab191394 was shown to react with Ikaros in wild-type Jurkat cells in western blot with loss of signal observed in IKZF1 knockout cell line ab274920 (knockout cell lysate **ab274978**). Wild-type and IKZF1 knockout Jurkat cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab191394** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 10000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Next Generation Sequencing - Human IKZF1 knockout Jurkat cell line (ab274920)

Knockout achieved by CRISPR/Cas9; X = 2 bp insertion, 1 bp insertion; Frameshift: 100%

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