abcam

Product datasheet

Human HNRNPA2B1 knockout HEK-293T cell line ab266404

4 Images

Overview

Product name Human HNRNPA2B1 knockout HEK-293T cell line

Parental Cell Line HEK293T
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, Homozygous: 2 bp deletion in exon 3

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

Tested applications Suitable for: WB

Biosafety level 2

General notesRecommended control: Human wild-type HEK293T cell line (<u>ab255449</u>). 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.

 $\textbf{Cryopreservation cell medium:} \ \ \textbf{Cell Freezing Medium-DMSO Serum free media, contains}$

8.7% DMSO in MEM supplemented with methyl cellulose.

Culture medium: DMEM (High Glucose) + 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 bath for 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 2x10⁴ cells/cm². 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% CO₂. 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 $2x10^4$ cells/cm² is recommended.

1

A partial media change 24 hours prior to subculture may be helpful to encourage growth, if

Cells should be passaged when they have achieved 80-90% confluence.

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We will provide viable cells that proliferate on revival.

Properties

Number of cells 1 x 10⁶ cells/vial, 1 mL

Adherent /Suspension Adherent
Tissue Kidney
Cell type epithelial

STR Analysis Amelogenin X D5S818: 8, 9 D13S317: 12, 14 D7S820: 11 D16S539: 9, 13 vWA: 16, 19 TH01:

7, 9.3 TPOX: 11 CSF1PO: 11, 12

Antibiotic resistance Puromycin 1.00µg/ml

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 Involved with pre-mRNA processing. Forms complexes (ribonucleosomes) with at least 20 other

different hnRNP and heterogeneous nuclear RNA in the nucleous.

Sequence similarities Contains 2 RRM (RNA recognition motif) domains.

Cellular localization Nucleus > nucleoplasm. Cytoplasm. Localized in cytoplasmic mRNP granules containing

untranslated mRNAs. Component of ribonucleosomes. Predominantly nucleoplasmic, however isoform A2 is also found in the cytoplasm of cells in some tissues. Not found in the nucleolus.

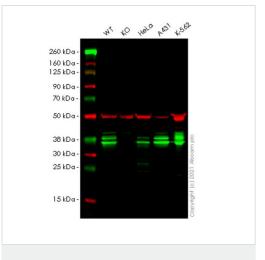
Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab266404 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. Predicted molecular weight: 37 kDa.

Images



Western blot - Human HNRNPA2B1 knockout HEK-293T cell line (ab266404)

All lanes : Anti-hnRNP A2B1 antibody [EPR24002-81] (**ab259894**) at 1/1000 dilution

Lane 1 : Wild-type HEK-293T (human embryonic kidney epithelial cell), whole cell lysate

Lane 2: hnRNP A2B1 knockout HEK-293T (human embryonic kidney epithelial cell), whole cell lysate

Lane 3 : HeLa (human cervix adenocarcinoma epithelial cell), whole cell lysate

Lane 4: A431 (human epidermoid carcinoma epithelial cell), whole cell lysate

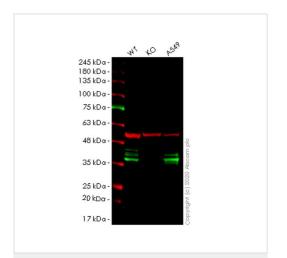
Lane 5: K562 (human chronic myelogenous leukemia lymphoblast), whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit IgG H&L (IRDye® 800CW)
(ab216773) and Goat Anti-Mouse IgG H&L (IRDye® 680RD)
(ab216776) at 1/10000 dilution

Predicted band size: 37 kDa **Observed band size:** 36, 38 kDa



Western blot - Human HNRNPA2B1 knockout HEK293T cell line (ab266404)

All lanes: Anti-hnRNP A2B1 antibody (ab31645) at 1/500 dilution

Lane 1: Wild-type HEK293T cell lysate

Lane 2: HNRNPA2B1 knockout HEK293T cell lysate

Lane 3: A549 cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 37 kDa **Observed band size:** 37 kDa

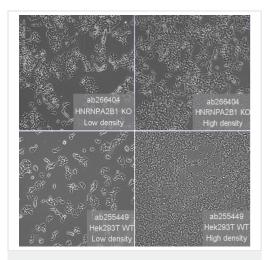
Lanes 1-3: Merged signal (red and green). Green - <u>ab31645</u> observed at 37 kDa. Red - loading control, <u>ab7291</u> observed at 50 kDa.

ab31645 Anti-hnRNP A2B1 antibody was shown to specifically react with hnRNP A2B1 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266404 (knockout cell lysate ab257224) was used. Wild-type and hnRNP A2B1 knockout samples were subjected to SDS-PAGE. ab31645 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) were incubated overnight at 4°C at 1 in 500 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.

Mut GGTAATGAGGGATCCTGCAAGCAAAAGA--AAGAGGATTTGGTTTTGTAACTTTTTCATC

WT GGTAATGAGGGATCCTGCAAGCAAAAGATCAAGAGGATTTGGTTTTGTAACTTTTTCATC

Sanger Sequencing - Human HNRNPA2B1 knockout HEK293T cell line (ab266404) Homozygous: 2 bp deletion in exon 3



Cell Culture - Human HNRNPA2B1 knockout HEK293T cell line (ab266404) Representative images of HNRNPA2B1 knockout HEK293T cells, low and high confluency examples (top left and right respectively) and wild-type HEK293T cells, low and high confluency (bottom left and right respectively) showing typical adherent, epithelial-like morphology. Images were captured at 10X magnification using a EVOS XL Core microscope.

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