abcam

Product datasheet

Human TLE1 knockout HeLa cell line ab264901

4 Images

Overview

Product name Human TLE1 knockout HeLa cell line

Parental Cell Line HeLa
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon 12 and 4 bp deletion in exon 12

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

Tested applications Suitable for: WB

Biosafety level

General notesRecommended control: Human wild-type HeLa cell line (<u>ab255448</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.

Cryopreservation cell medium: 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.

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

1

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

This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our limited use license and patent pages.

We will provide viable cells that proliferate on revival.

Properties

Cell type

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

Adherent/Suspension Adherent **Tissue** Cervix

Disease Adenocarcinoma

Gender Female

STR Analysis Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18

TH01: 7 TPOX: 8, 12 CSF1PO: 9, 10

Antibiotic resistance Puromycin 1.00µg/ml

Mycoplasma free Yes

Storage instructions Shipped on Dry Ice. Store in liquid nitrogen.

epithelial

Storage buffer Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

Target

Function Transcriptional corepressor that binds to a number of transcription factors. Inhibits NF-kappa-B-

> regulated gene expression. Inhibits the transcriptional activation mediated by FOXA2, and by CTNNB1 and TCF family members in Wnt signaling. The effects of full-length TLE family members may be modulated by association with dominant-negative AES. Unusual function as coactivator

for ESRRG.

Tissue specificity In all tissues examined, mostly in brain, liver and muscle.

Sequence similarities Belongs to the WD repeat Groucho/TLE family.

Contains 6 WD repeats.

Domain WD repeat Groucho/TLE family members are characterized by 5 regions, a glutamine-rich Q

> domain, a glycine/proline-rich GP domain, a central CcN domain, containing a nuclear localization signal, and a serine/proline-rich SP domain. The most highly conserved are the N-terminal Q

domain and the C-terminal WD-repeat domain.

Post-translational

Phosphorylated, probably by CDK1. The degree of phosphorylation varies throughout the cell modifications cycle, and is highest at the G2/M transition. Becomes hyperphosphorylated in response to cell

differentiation and interaction with HES1 or RUNX1.

Ubiquitinated by XIAP/BIRC4.

Cellular localization Nucleus. Nuclear and chromatin-associated, depending on isoforms and phosphorylation status.

Hyperphosphorylation decreases the affinity for nuclear components.

Applications

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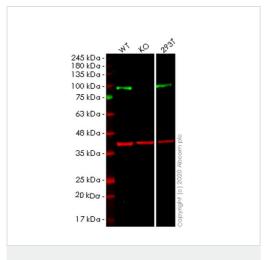
The Abpromise guarantee

Our Abpromise guarantee covers the use of ab264901 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: 83 kDa.

Images



Western blot - Human TLE1 knockout HeLa cell line (ab264901)

All lanes : Anti-TLE 1 antibody [EPR9386(2)] (<u>ab183742</u>) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: TLE1 knockout HeLa cell lysate

Lane 3: 293T cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 83 kDa Observed band size: 83 kDa

Lanes 1-3: Merged signal (red and green). Green - <u>ab183742</u> observed at 83 kDa. Red - loading control, <u>ab8245</u> observed at 37 kDa.

<u>ab183742</u> Anti-TLE 1 antibody [EPR9386(2)] was shown to specifically react with TLE 1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab264901 (knockout cell lysate <u>ab257241</u>) was used. Wild-type and TLE 1 knockout samples were subjected to SDS-PAGE. <u>ab183742</u> and Anti-GAPDH antibody [EPR16891] - Loading Control (<u>ab8245</u>) 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 (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye®680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.

Mut AGCCTCCAGCCATAGACCCCCTCGTTAACC----GGGTACAGCATCGCCTCTTGTTGTGC

Sanger Sequencing - Human TLE1 knockout HeLa cell line (ab264901)

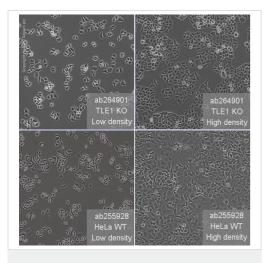
Allele-1: 4 bp deletion in exon 12.

Mut AGCCTCCAGCCATAGACCCCCTCGTTAACC-AGCGGGTACAGCATCGCCTCTTGTTGTGC

WT AGCCTCCAGCCATAGACCCCCTCGTTAACCAAGCGGGTACAGCATCGCCTCTTGTTGTGC

Sanger Sequencing - Human TLE1 knockout HeLa cell line (ab264901)

Allele-2: 1 bp deletion in exon 12.



Cell Culture - Human TLE1 knockout HeLa cell line (ab264901)

Representative images of TLE1 knockout HeLa cells, low and high confluency examples (top left and right respectively) and wild-type HeLa 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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