abcam

Product datasheet

Human RBBP7 (RbAp46) knockout HeLa cell line ab264677

4 Images

Overview

Product name	Human RBBP7 (RbAp46) knockout HeLa cell line
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon 2 and 4 bp deletion in exon 2 and Insertion of the selection cassette in exon 2
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Tested applications	**Suitable for:** WB
Biosafety level	2
General notes	**Recommended control:** Human wild-type HeLa cell line (**ab255448**). 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 $2x10^4$ cells/cm^2. 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 $2x10^4$ cells/cm^2 is recommended.
A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required.
Cells should be passaged when they have achieved 80-90% confluence.
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	Adherent
Tissue	Cervix
Cell type	epithelial
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
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	Core histone-binding subunit that may target chromatin remodeling factors, histone acetyltransferases and histone deacetylases to their histone substrates in a manner that is regulated by nucleosomal DNA. Component of several complexes which regulate chromatin metabolism. These include the type B histone acetyltransferase (HAT) complex, which is required for chromatin assembly following DNA replication; the core histone deacetylase (HDAC) complex, which promotes histone deacetylation and consequent transcriptional repression; the nucleosome remodeling and histone deacetylase complex (the NuRD complex), which promotes transcriptional repression by histone deacetylation and nucleosome remodeling; and the PRC2/EED-EZH2 complex, which promotes repression of homeotic genes during development; and the NURF (nucleosome remodeling factor) complex.
Sequence similarities	Belongs to the WD repeat RBAP46/RBAP48/MSI1 family. Contains 7 WD repeats.
Cellular localization	Nucleus.

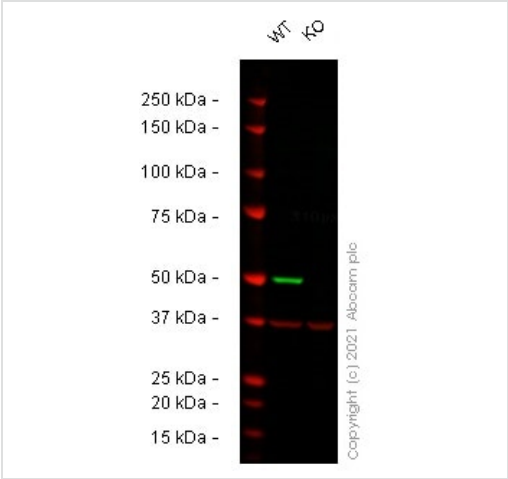
Applications

The Abpromise guarantee	Our **Abpromise guarantee** covers the use of ab264677 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: 48 kDa.

Images



Western blot - Human RBBP7 (RbAp46) knockout HeLa cell line (ab264677)

All lanes : Anti-RBBP7 antibody [EPR23796-74] - ChIP Grade (**ab259957**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : RBBP7 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 48 kDa
Observed band size: 50 kDa

Lanes 1 - 2: Merged signal (red and green). Green - **ab259957** observed at 50 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab259957 was shown to react with RBBP7 in wild-type HeLa cells in Western blot with loss of signal observed in RBBP7 knockout cell line ab264677 (RBBP7 knockout cell lysate **ab258628**). Wild-type HeLa and RBBP7 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab259957** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 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 h at room temperature before imaging.

Sanger Sequencing - Human RBBP7 knockout HeLa cell line (ab264677)

Allele-2: 1 bp deletion in exon 2.

Sanger Sequencing - Human RBBP7 knockout HeLa cell line (ab264677)

Allele-1: 4 bp deletion in exon 2.

Sanger Sequencing - Human RBBP7 knockout HeLa cell line (ab264677)

Allele-3: Insertion of the selection cassette in exon 2.

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