abcam

Product datasheet

Human BECN1 (Beclin 1) knockout HeLa cell line ab262511

2 Images

Overview

Product name	Human BECN1 (Beclin 1) knockout HeLa cell line
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by CRISPR/Cas9; X = 4 bp deletion; Frameshift: 93.41%
Passage number	<20
Knockout validation	Next Generation Sequencing (NGS), Western Blot (WB)
Tested applications	**Suitable for:** WB
Biosafety level	2
General notes	**Recommended control:** Human wild-type HeLa cell line (**ab271142**). 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.
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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
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	Plays a central role in autophagy (By similarity). May play a role in antiviral host defense. Protects against infection by a neurovirulent strain of Sindbis virus.
Tissue specificity	Ubiquitous.
Sequence similarities	Belongs to the beclin family.
Cellular localization	Golgi apparatus > trans-Golgi network membrane. Interaction with ATG14 promotes translocation to autophagosomes. Expressed in dendrites and cell bodies of cerebellar Purkinje cells.

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab262511 in the following tested applications.

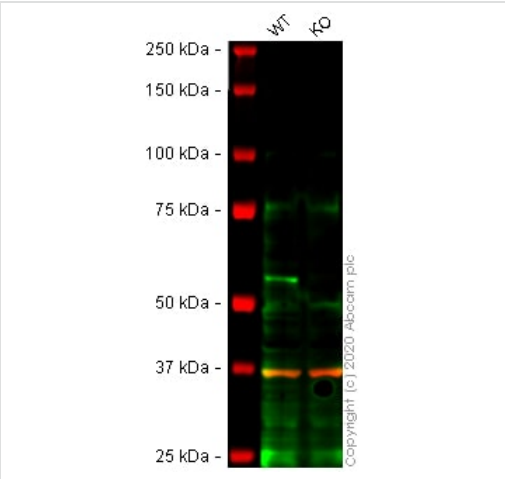
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration.

Images

Next Generation Sequencing - Human BECN1 (Beclin 1) knockout HeLa cell line (ab262511)

4 bp deletion after Gln74 of the WT protein



Western blot - Human BECN1 (Beclin 1) knockout HeLa cell line (ab262511)

All lanes : Anti-Beclin 1 antibody [OTI4A10] (**ab118148**) at 1/1000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : BECN1 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Lanes 1 - 2: Merged signal (red and green). Green - **ab118148** observed at 60 kDa. Red - loading control, **ab181602** (Rabbit Anti-GAPDH antibody [EPR16891]) observed at 37kDa.

ab118148 was shown to react with Beclin 1 in wild-type HeLa cells in western blot Loss of signal was observed when BECN1 knockout cell line ab262511 (knockout cell lysate **ab263936**) was used. Wild-type HeLa and BECN1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in non-mammalian (TBS-based) blocking solution before incubation with **ab118148** and **ab181602** (Rabbit Anti-GAPDH antibody [EPR16891]) 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 hour at room temperature before imaging.

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