

Human CEACAM1 knockout A549 cell line ab267046

5 Images

Overview

Product name	Human CEACAM1 knockout A549 cell line
Parental Cell Line	A549
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp insertion in exon 3 and 2 bp insertion in exon 3 and 5 bp deletion in exon 3
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Tested applications	Suitable for: WB
Biosafety level	2
General notes	<p>Recommended control: Human wild-type A549 cell line (ab255450). 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.</p> <p>Cryopreservation cell medium: Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p>Culture medium: F-12K + 10% FBS</p> <p>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.</p> <ol style="list-style-type: none"> 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 2×10^3-1×10^4 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. <p>Subculture guidelines:</p> <p>All seeding densities should be based on cell counts gained by established methods. A guide seeding density of 6×10^4 cells/cm² is recommended.</p> <p>A partial media change 24 hours prior to subculture may be helpful to encourage growth, if</p>

required.

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

Do not exceed 7×10^4 cells/cm².

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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	Lung
Cell type	epithelial
Disease	Carcinoma
Gender	Male
STR Analysis	Amelogenin X,YD5S818: 11 D13S317: 11 D7S820: 8, 11 D16S539: 11, 12 WWA: 14 TH01: 8,9,3 TPOX: 8,11 CSF1PO: 10, 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

Sequence similarities	Belongs to the immunoglobulin superfamily. CEA family. Contains 3 Ig-like C2-type (immunoglobulin-like) domains. Contains 1 Ig-like V-type (immunoglobulin-like) domain.
Cellular localization	Secreted; Cell membrane and Cell membrane. Localizes to sites of cell-cell contact.
Form	There are 11 isoforms produced by alternative splicing. Isoform 1 = BGP _a ; CEACAM1-4L; TM1-CEA; Isoform 2 = BGP _g ; CEACAM1-4C1; Isoform 3 = BGP _h ; CEACAM1-3; Isoform 4 = BGP _i ; CEACAM1-3C2; Isoform 5 = BGP _y ; CEACAM1-3AL; Isoform 6 = BGP _b ; CEACAM1-3L; TM2-CEA; Isoform 7 = BGP _x ; CEACAM1-1L; Isoform 8 = BGP _c ; CEACAM1-4S; TM3-CEA; Isoform 9 = BGP _z ; CEACAM1-3AS and Isoform 11 = BGP _d ; CEACAM1-3S.

Applications

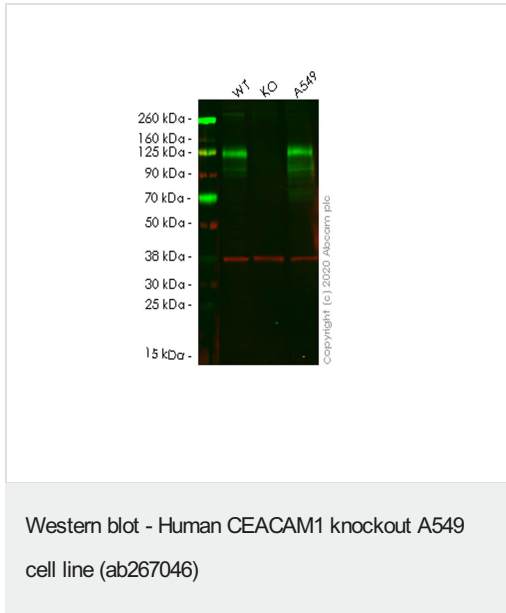
The Abpromise guarantee Our [Abpromise guarantee](#) covers the use of ab267046 in the following tested applications.

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

Application	Abreviews	Notes

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WB		Use at an assay dependent concentration. Predicted molecular weight: 57 kDa.

Images



All lanes : Anti-CEACAM1 antibody [EPR4049] ([ab108397](#)) at 1/1000 dilution

Lane 1 : Wild-type A549 cell lysate

Lane 2 : CEACAM1 knockout A549 cell lysate

Lane 3 : A549 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#)) at 1/10000 dilution

Predicted band size: 57 kDa

Observed band size: 100-180 kDa

Lanes 1-4: Merged signal (red and green). Green - [ab108397](#) observed at 100-180 kDa. Red - loading control [ab8245](#) observed at 36 kDa.

[ab108397](#) Anti-CEACAM1 antibody [EPR4049] was shown to specifically react with CEACAM1 in wild-type A549 cells. Loss of signal was observed when knockout cell line ab267046 (knockout cell lysate [ab257388](#)) was used. Wild-type and CEACAM1 knockout samples were subjected to SDS-PAGE. [ab108397](#) and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed ([ab216776](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

```

Mut  CAGGTGAAGGCCACAGCATCCTTGTCTCC- - - - GTTGGAGTTGTTGCTGGAGATGGAG
      |||
WT   CAGGTGAAGGCCACAGCATCCTTGTCTCCACAGGGTTGGAGTTGTTGCTGGAGATGGAG

```

Sanger Sequencing - Human CEACAM1 knockout
A549 cell line (ab267046)

Allele-1: 5 bp deletion in exon3

```

Mut  CAGGTGAAGGCCACAGCATCCTTGTCTCCACAGGGTTGGAGTTGTTGCTGGAGATGGA
      |||
WT   CAGGTGAAGGCCACAGCATCCTTGTCTCC ACAGGGTTGGAGTTGTTGCTGGAGATGGA

```

Sanger Sequencing - Human CEACAM1 knockout
A549 cell line (ab267046)

Allele-2: 1 bp insertion in exon 3.

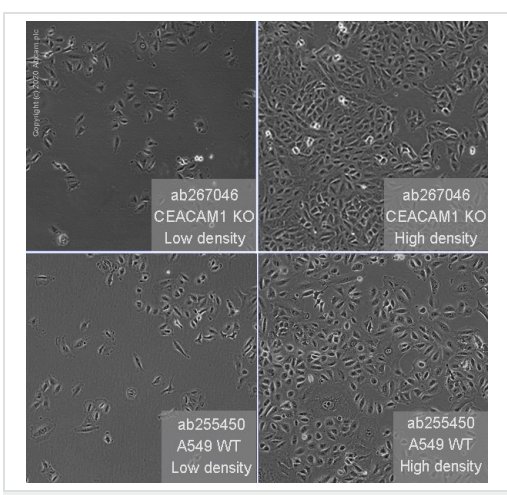
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Mut  CAGGTGAAGGCCACAGCATCCTTGTCTCCACAGGGTTGGAGTTGTTGCTGGAGATGG
      |||
WT   CAGGTGAAGGCCACAGCATCCTTGTCTCC ACAGGGTTGGAGTTGTTGCTGGAGATGG

```

Sanger Sequencing - Human CEACAM1 knockout
A549 cell line (ab267046)

Allele-3: 2 bp insertion in exon 3.



Representative images of CEACAM1 knockout A549 cells, low and high confluency examples (top left and right respectively) and wild-type A549 cells, low and high confluency (bottom left and right respectively) showing typical adherent, epithelial-like morphology. Images were captured at 10X magnification using an EVOS M5000 microscope.

Cell Culture - Human CEACAM1 knockout A549 cell line (ab267046)

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