abcam

Product datasheet

Human CPS1 knockout HeLa cell line ab261809

4 Images

Overview

Product name	Human CPS1 knockout HeLa cell line
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp insertion in exon 15 and Insertion of the selection cassette in exon 15
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 (**ab255928**). 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
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	Involved in the urea cycle of ureotelic animals where the enzyme plays an important role in removing excess ammonia from the cell.
Tissue specificity	Primarily in the liver and small intestine.
Involvement in disease	Defects in CPS1 are the cause of carbamoyl phosphate synthetase 1 deficiency (CPS1D) [MIM:237300]. CPS1D is an autosomal recessive disorder of the urea cycle causing hyperammonemia. Clinical features include protein intolerance, intermittent ataxia, seizures, lethargy, developmental delay and mental retardation. Note=Genetic variations in CPS1 influence the availability of precursors for nitric oxide (NO) synthesis and play a role in clinical situations where endogenous NO production is critically important, such as neonatal pulmonary hypertension, increased pulmonary artery pressure following surgical repair of congenital heart defects or hepatovenocclusive disease following bone marrow transplantation. Infants with neonatal pulmonary hypertension homozygous for Thr-1406 have lower L-arginine concentrations than neonates homozygous for Asn-1406.
Sequence similarities	Contains 2 ATP-grasp domains. Contains 1 glutamine amidotransferase type-1 domain.
Domain	The type-1 glutamine amidotransferase domain is defective.
Cellular localization	Mitochondrion.

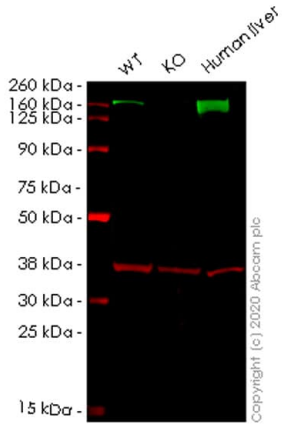
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab261809 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: 165 kDa.

Images



Western blot - Human CPS1 knockout HeLa cell line (ab261809)

All lanes : Anti-CPS1 antibody [EPR7493-29] (**ab155083**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : CPS1 knockout HeLa cell lysate
Lane 3 : Human liver tissue lysate

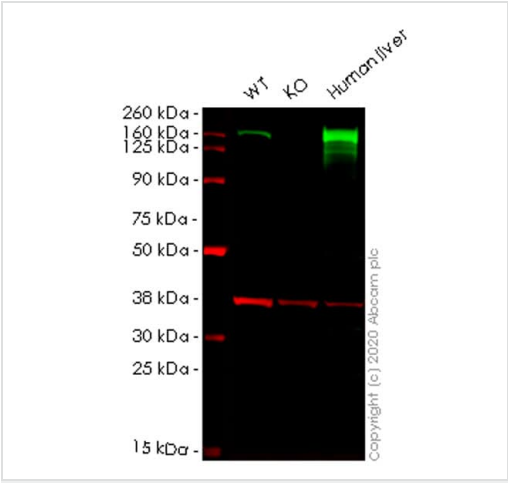
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 165 kDa
Observed band size: 165 kDa

Lanes 1-3: Merged signal (red and green). Green - **ab155083** observed at 165 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab155083 Anti-CPS1 antibody [EPR7493-29] was shown to specifically react with CPS1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab261809 (knockout cell lysate **ab257121**) was used. Wild-type and CPS1 knockout samples were subjected to SDS-PAGE. **ab155083** 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 10000 dilution for 1 hour at room temperature before imaging.



Western blot - Human CPS1 knockout HeLa cell line (ab261809)

All lanes : Anti-CPS1 antibody [EPR7493-3] (**ab129076**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : CPS1 knockout HeLa cell lysate
Lane 3 : Human liver tissue lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 165 kDa
Observed band size: 165 kDa

Lanes 1-3: Merged signal (red and green). Green - **ab129076** observed at 165 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab129076 Anti-CPS1 antibody [EPR7493-3] was shown to specifically react with CPS1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab261809 (knockout cell lysate **ab257121**) was used. Wild-type and CPS1 knockout samples were subjected to SDS-PAGE. **ab129076** 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 10000 dilution for 1 hour at room temperature before imaging.

Sanger Sequencing - Human CPS1 knockout HeLa cell line (ab261809)

Allele-1: 1 bp insertion in exon 15.

Sanger Sequencing - Human CPS1 knockout HeLa cell line (ab261809)

Allele-2: Insertion of the selection cassette in exon 15.

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