abcam

Product datasheet

Human ACE2 knockout Hep G2 cell line ab273733

画像数 4

製品の概要

製品名	Human ACE2 knockout Hep G2 cell line
Parental Cell Line	HepG2
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 71 bp deletion in exon 2
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
アプリケーション	**適用あり:** WB
Biosafety level	1
特記事項	**Recommended control:** Human wild-type HepG2 cell line (**ab275467**). 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:** MEM + 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 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.

製品の特性

Number of cells	1 x 10^6 cells/vial, 1 mL
Adherent /Suspension	Adherent
Tissue	Liver
Cell type	epithelial
Disease	Hepatocellular Carcinoma
Gender	Male
Mycoplasma free	Yes
保存方法	Shipped on Dry Ice. Store in liquid nitrogen.
バッファー	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

ターゲット情報

機能	Carboxypeptidase which converts angiotensin I to angiotensin 1-9, a peptide of unknown function, and angiotensin II to angiotensin 1-7, a vasodilator. Also able to hydrolyze apelin-13 and dynorphin-13 with high efficiency. May be an important regulator of heart function. In case of human coronaviruses SARS and HCoV-NL63 infections, serve as functional receptor for the spike glycoprotein of both coronaviruses.
組織特異性	Expressed in endothelial cells from small and large arteries, and in arterial smooth muscle cells. Expressed in lung alveolar epithelial cells, enterocytes of the small intestine, Leydig cells and Sertoli cells (at protein level). Expressed in heart, kidney, testis, and gastrointestinal system.
配列類似性	Belongs to the peptidase M2 family.
翻訳後修飾	N-glycosylation on Asn-90 may limit SARS infectivity.
細胞内局在	Secreted and Cell membrane.

アプリケーション

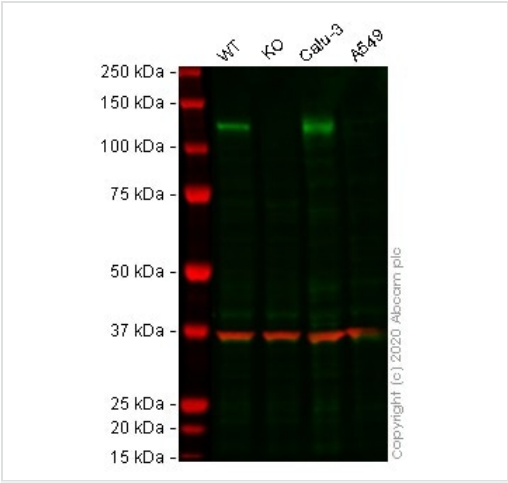
The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab273733の使用に適用されます

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 92 kDa.

画像

画像



Western blot - Human ACE2 knockout HepG2 cell line (ab273733)

All lanes : Anti-ACE2 antibody [EPR4435(2)] (**ab108252**) at 1/1000 dilution

Lane 1 : Wild-type HepG2 cell lysate
Lane 2 : ACE2 knockout HepG2 cell lysate
Lane 3 : Calu-3 cell lysate
Lane 4 : A549 cell lysate

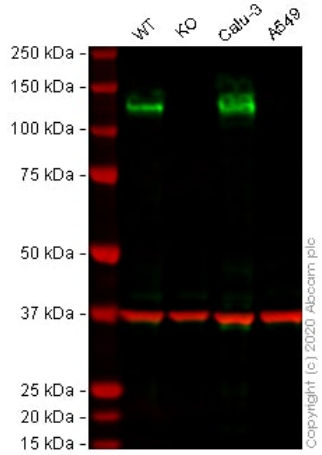
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

Predicted band size: 92 kDa
Observed band size: 130 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab108252** observed at 130 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab108252 was shown to react with ACE2 in wild-type HepG2 cells in western blot with loss of signal observed in ACE2 knockout cell line ab273733 (knockout cell lysate **ab275495**). Wild-type and ACE2 knockout HepG2 cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab108252** 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 hour at room temperature before imaging.



Western blot - Human ACE2 knockout HepG2 cell line (ab273733)

All lanes : Anti-ACE2 antibody [EPR4436] (**ab108209**) at 1/1000 dilution

Lane 1 : Wild-type HepG2 cell lysate
Lane 2 : ACE2 knockout HepG2 cell lysate
Lane 3 : Calu-3 cell lysate
Lane 4 : A549 cell lysate

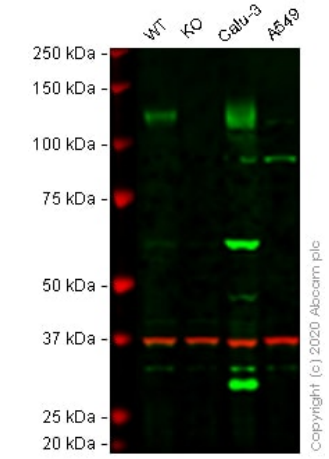
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

Predicted band size: 92 kDa
Observed band size: 130 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab108209** observed at 130 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab108209 was shown to react with ACE2 in wild-type HepG2 cells in western blot with loss of signal observed in ACE2 knockout cell line ab273733 (knockout cell lysate **ab275495**). Wild-type and ACE2 knockout HepG2 cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab108209** 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 hour at room temperature before imaging.



Western blot - Human ACE2 knockout HepG2 cell line (ab273733)

All lanes : Anti-ACE2 antibody (**ab65863**) at 1 µg/ml

Lane 1 : Wild-type HepG2 cell lysate
Lane 2 : ACE2 knockout HepG2 cell lysate
Lane 3 : Calu-3 cell lysate
Lane 4 : A549 cell lysate

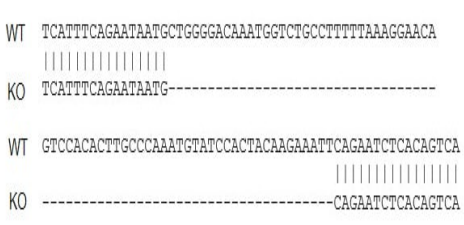
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

Predicted band size: 92 kDa
Observed band size: 130 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab65863** observed at 130 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab65863 was shown to react with ACE2 in wild-type HepG2 cells in western blot with loss of signal observed in ACE2 knockout cell line ab273733 (knockout cell lysate **ab275495**). Wild-type and ACE2 knockout HepG2 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab65863** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at 1 µg/ml 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.



Sanger Sequencing - Human ACE2 knockout HepG2 cell line (ab273733)

Allele 1: 71 bp deletion in exon 2.

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