abcam

Product datasheet

Human POR (Cytochrome P450 Reductase) knockout HeLa cell line ab264996

画像数 2

製品の概要

製品名	Human POR (Cytochrome P450 Reductase) knockout HeLa cell line
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: Insertion of the selection cassette in exon 4
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	2
特記事項	**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.
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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	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
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	This enzyme is required for electron transfer from NADP to cytochrome P450 in microsomes. It can also provide electron transfer to heme oxygenase and cytochrome B5.
関連疾患	Defects in POR are the cause of adrenal hyperplasia variant type (AHV) [MIM:201750]; also known as Antley-Bixler syndrome-like phenotype with disordered steroidogenesis. AHV is a rare variant of congenital adrenal hyperplasia. It is an autosomal recessive disorder with apparent combined P450C17 and P450C21 deficiency. Affected girls are born with ambiguous genitalia, but their circulating androgens are low and virilization does not progress. Conversely, affected boys are sometimes born undermasculinized. Boys and girls can also present with bone malformations, in some cases resembling the pattern seen in patients with Antley-Bixler syndrome. Defects in POR are a cause of isolated disordered steroidogenesis (IDS) [MIM:201750].
配列類似性	In the C-terminal section; belongs to the flavoprotein pyridine nucleotide cytochrome reductase family. Contains 1 FAD-binding FR-type domain. Contains 1 flavodoxin-like domain.
細胞内局在	Endoplasmic reticulum membrane. Anchored to the ER membrane by its N-terminal hydrophobic region.

アプリケーション

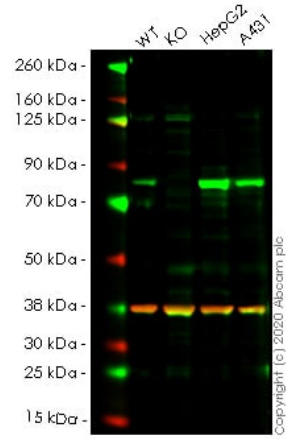
アプリケーション

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ab264996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: 76 kDa.

画像



Western blot - Human POR knockout HeLa cell line (ab264996)

All lanes : Anti-Cytochrome P450 Reductase antibody [EPR14479(B)] (**ab180597**) at 1/10000 dilution

Lane 1 : Wild-type HeLa lysate
Lane 2 : Cytochrome P450 Reductase knockout HeLa lysate
Lane 3 : HepG2 lysate
Lane 4 : A431 lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 76 kDa

Lanes 1-4: Merged signal (red and green). Green - **ab180597** observed at 75 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab180597 Anti-Cytochrome P450 Reductase antibody [EPR14479(B)] was shown to specifically react with Cytochrome P450 Reductase in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab264996 (knockout cell lysate **ab257595**) was used. Wild-type and Cytochrome P450 Reductase knockout samples were subjected to SDS-PAGE. **ab180597** and Anti-GAPDH antibody [6C5] - Loading Control?(**ab8245**) were incubated overnight at 4^°C at 1 in 10000 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.

Sanger Sequencing - Human POR knockout HeLa cell line (ab264996)

Homozygous: Insertion of the selection cassette in exon 4.

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