abcam

Product datasheet

Human UCHL1 (PGP9.5) knockout HEK-293T cell line ab255443

画像数 2

製品の概要

製品名 Human UCHL1 (PGP9.5) knockout HEK-293T cell line

Parental Cell Line HEK293T
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, Homozygous: 45 bp deletion in exon 1

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

2

アプリケーション **適用あり**: WB

Biosafety level

特記事項 Recommended control: Human wild-type HEK293T cell line (<u>ab255593</u>). 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 2x10⁴ 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.

Subculture guidelines:

All seeding densities should be based on cell counts gained by established methods. A guide seeding density of $2x10^4$ cells/cm² is recommended.

1

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 10⁶ cells/vial, 1 mL

Adherent /Suspension Adherent
Tissue Kidney
Cell type epithelial

STR Analysis Amelogenin X D5S818: 8, 9 D13S317: 11, 12, 14 D7S820: 11 D16S539: 9, 13 vWA: 15, 20

TH01: 7, 9.3 TPOX: 11, 12 CSF1PO: 12

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 Ubiquitin-protein hydrolase involved both in the processing of ubiquitin precursors and of

ubiquitinated proteins. This enzyme is a thiol protease that recognizes and hydrolyzes a peptide bond at the C-terminal glycine of ubiquitin. Also binds to free monoubiquitin and may prevent its degradation in lysosomes. The homodimer may have ATP-independent ubiquitin ligase activity.

組織特異性 Found in neuronal cell bodies and processes throughout the neocortex (at protein level).

Expressed in neurons and cells of the diffuse neuroendocrine system and their tumors. Weakly expressed in ovary. Down-regulated in brains from Parkinson disease and Alzheimer disease

patients.

関連疾患 Parkinson disease 5

Neurodegeneration with optic atrophy, childhood-onset

配列類似性 Belongs to the peptidase C12 family.

翻訳後修飾 O-glycosylated.

細胞内局在 Cytoplasm. Endoplasmic reticulum membrane. About 30% of total UCHL1 is associated with

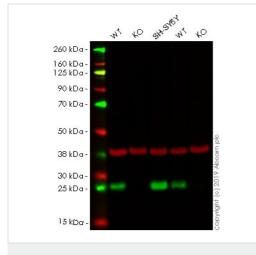
membranes in brain.

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab255443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: 24 kDa.

画像



Western blot - Human UCHL1 (PGP9.5) knockout HEK-293T cell line (ab255443) **All lanes :** Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (**ab108986**) at 1/1000 dilution

Lane 1: Wild-type HAP1 cell lysate

Lane 2: UCHL1 knockout HAP1 cell lysate

Lane 3: SH-SY5Y cell lysate

Lane 4: Wild-type HEK-293T cell lysate

Lane 5: UCHL1 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 24 kDa

Additional bands at: 37 kDa (possible Loading Control)

Lanes 1 - 5: Merged signal (red and green). Green - <u>ab108986</u> observed at 25 kDa. Red - loading control, <u>ab8245</u> observed at 37 kDa.

ab108986 was shown to react with PGP9.5 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab255443 (knockout cell lysate ab263773) was used. Wild-type and PGP9.5 knockout samples were subjected to SDS-PAGE. ab108986 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.



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