abcam

Product datasheet

Human PRDX1 (Peroxiredoxin 1/PAG) knockout HEK-293T cell line ab266842

画像数 5

製品の概要

製品名	Human PRDX1 (Peroxiredoxin 1/PAG) knockout HEK-293T cell line
Parental Cell Line	HEK293T
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 1 bp deletion in exon 2
Passage number	<20
Knockout validation	Immunocytochemistry (ICC), Sanger Sequencing, Western Blot (WB)
アプリケーション	**適用あり:** WB, ICC/IF
Biosafety level	2
特記事項	**Recommended control:** Human wild-type HEK293T cell line (**ab255449**). 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.
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	Kidney
Cell type	epithelial
STR Analysis	Amelogenin X D5S818: 8, 9 D13S317: 12, 14 D7S820: 11 D16S539: 9, 13 vWA: 16, 19 TH01: 7, 9.3 TPOX: 11 CSF1PO: 11, 12
Antibiotic resistance	Puromycin 1.00µg/ml
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	Involved in redox regulation of the cell. Reduces peroxides with reducing equivalents provided through the thioredoxin system but not from glutaredoxin. May play an important role in eliminating peroxides generated during metabolism. Might participate in the signaling cascades of growth factors and tumor necrosis factor-alpha by regulating the intracellular concentrations of H(2)O(2). Reduces an intramolecular disulfide bond in GDPD5 that gates the ability to GDPD5 to drive postmitotic motor neuron differentiation.
配列類似性	Belongs to the ahpC/TSA family. Contains 1 thioredoxin domain.
翻訳後修飾	Phosphorylated on Thr-90 during the M-phase, which leads to a more than 80% decrease in enzymatic activity.
細胞内局在	Cytoplasm. Melanosome. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

アプリケーション

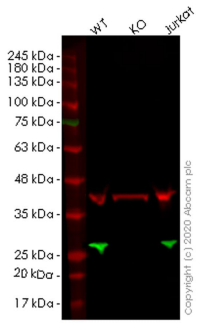
The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab266842の使用に適用されます

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

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 22 kDa.
ICC/IF		Use at an assay dependent concentration.

画像



Western blot - Human PRDX1 knockout HEK293T cell line (ab266842)

All lanes : Anti-Peroxiredoxin 1/PAG antibody [EPR5434] (**ab109506**) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : PRDX1 knockout HEK293T cell lysate
Lane 3 : Jurkat cell lysate

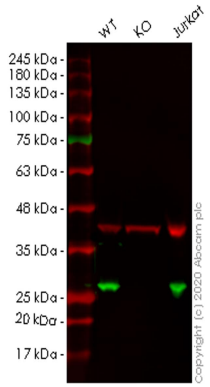
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 22 kDa
Observed band size: 26 kDa

Lanes 1-3: Merged signal (red and green). Green - **ab109506** observed at 26 kDa. Red - loading control **ab8245** observed at 36 kDa.

ab109506 Anti-Peroxiredoxin 1/PAG antibody [EPR5434] was shown to specifically react with Peroxiredoxin 1/PAG in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266842 (knockout cell lysate **ab257040**) was used. Wild-type and Peroxiredoxin 1/PAG knockout samples were subjected to SDS-PAGE. **ab109506** 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.



Western blot - Human PRDX1 knockout HEK293T cell line (ab266842)

All lanes : Anti-Peroxiredoxin 1/PAG antibody [EPR5433] (**ab109498**) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : PRDX1 knockout HEK293T cell lysate
Lane 3 : Jurkat cell lysate

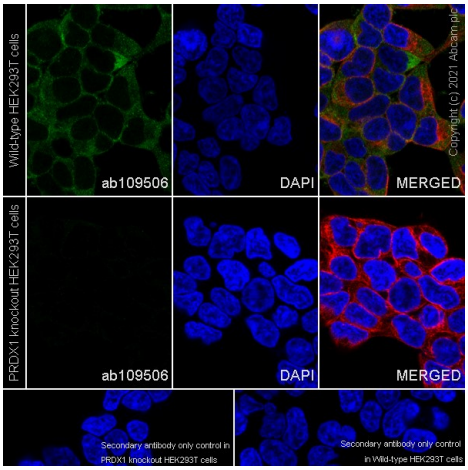
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 22 kDa
Observed band size: 26 kDa

Lanes 1-3: Merged signal (red and green). Green - **ab109498** observed at 26 kDa. Red - loading control **ab8245** observed at 36 kDa.

ab109498 Anti-Peroxiredoxin 1/PAG antibody [EPR5433] was shown to specifically react with Peroxiredoxin 1/PAG in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266842 (knockout cell lysate **ab257040**) was used. Wild-type and Peroxiredoxin 1/PAG knockout samples were subjected to SDS-PAGE. **ab109498** 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.



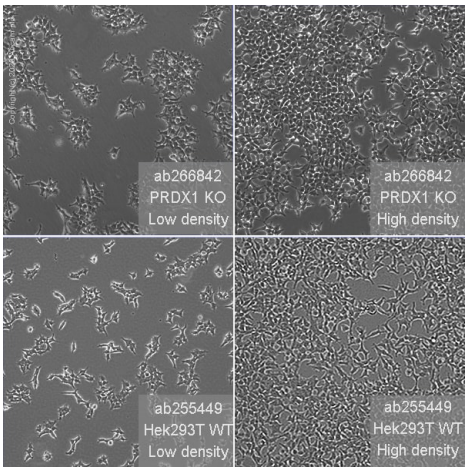
Immunocytochemistry/ Immunofluorescence - Human PRDX1 (Peroxiredoxin 1/PAG) knockout HEK-293T cell line (ab266842)

Peroxiredoxin 1/PAG (PRDX1) staining observed in wild-type HEK293T cells and PRDX1 knockout HEK293T cells (ab266842). The cells were fixed with 100% methanol then permeabilized with 0.1% Triton X-100. The cells were then incubated with **ab109506** at 1/50 dilution and followed by secondary antibody **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution (shown in green). **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain at 1/200 dilution (shown in red). Nuclear DNA was labelled in blue with DAPI.

Confocal image showing cytoplasmic staining in wild-type HEK-293Tcell line, and no staining in PRDX1 knockout HEK-293T cell line.

Sanger Sequencing - Human PRDX1 knockout HEK293T cell line (ab266842)

Homozygous: 1 bp deletion in exon2



Cell Culture - Human PRDX1 (Peroxiredoxin 1/PAG) knockout HEK293T cell line (ab266842)

Representative images of PRDX1 knockout HEK293T cells, low and high confluency examples (top left and right respectively) and wild-type HEK293T 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.

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