abcam

Product datasheet

Human PPIF (Cyclophilin F) knockout HEK-293T cell lysate ab257039

3 Images

Overview

Product name Human PPIF (Cyclophilin F) knockout HEK-293T cell lysate

Product overview

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line HEK293T
Organism Human

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

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

Reconstitution notesTo use as WB control, resuspend the lyophilizate in 50 μL of LDS* Sample Buffer to have a final

concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M

DTT.

*Usage of SDS sample buffer is not recommended with these lyophilized lysates.

Notes

Lysate preparation: Our lysates are made using RIPA buffer to which we add a protease

inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found **here**. Please refer to our lysis protocol for further details on how our lysates are

prepared.

User storage instructions: Lyophilizate may be stored at 4°C. After reconstitution, store at -

20°C for short-term storage or -80°C for long-term storage.

Access thousands of knockout cell lysates, generated from commonly used cancer cell lines.

See here for more information on knockout cell lysates.

Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of

products that contain European Authorisation list (Annex XIV) substances.

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licenses and patents please refer to our $\underline{\text{limited use license}}$ and $\underline{\text{patent pages}}$.

Tested applications Suitable for: WB

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Properties

Storage instructions Store at

Store at -80°C. Please refer to protocols.

| Components | 1 kit |
|--|-----------|
| ab260168 - Human PPIF knockout HEK293T cell lysate | 1 x 100μg |
| ab255553 - Human wild-type HEK293T cell lysate | 1 x 100μg |

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

Target

Function PPlases accelerate the folding of proteins. It catalyzes the cis-trans isomerization of proline imidic

peptide bonds in oligopeptides.

Sequence similarities Belongs to the cyclophilin-type PPlase family.

Contains 1 PPlase cyclophilin-type domain.

Cellular localization Mitochondrion matrix.

Form This gene encodes a 178 aa mature protein that is found in the mitochondrion and may

participate in the permeability transition pore. While technically this protein is Cyclophilin F, literature references commonly refer to this protein as 'cyclophilin D' or 'CypD'. A different cytoplasmic protein of 370 aa, represented by Entrez GenelD 5481, is identified as Cyclophilin D.

This antibody does not react with this 370 aa cytoplasmic protein.

Applications

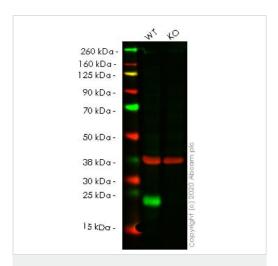
The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab257039 in the following tested applications.

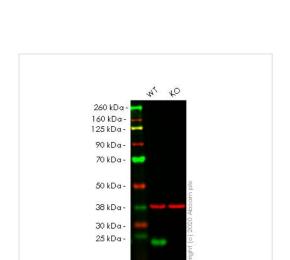
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

| Application | Abreviews | Notes |
|-------------|-----------|--|
| WB | | Use at an assay dependent concentration. Predicted molecular weight: 22 kDa. |

Images



Western blot - Human PPIF (Cyclophilin F) knockout HEK293T cell Iysate (ab257039)



Western blot - Human PPIF (Cyclophilin F) knockout HEK293T cell lysate (ab257039)

15 kDa

Lane 1: Wild-type HEK-293T cell lysate (20µg)

Lane 2: PPIF knockout HEK-293T cell lysate (20µg)

Lanes 1-2: Merged signal (red and green). Green - <u>ab110324</u> observed at 23 kDa. Red - loading control <u>ab181602</u> observed at 37 kDa.

ab110324 Anti-Cyclophilin F antibody [E11AE12BD4] was shown to specifically react with Cyclophilin F in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266077 (knockout cell lysate ab257039) was used. Wild-type and Cyclophilin F knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab110324 and Anti-GAPDH antibody[EPR16891] - Loading Control (ab181602) were incubated overnight at 4°C at 1 in 10000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Lane 1: Wild-type HEK-293T cell lysate (20µg)

Lane 2: PPIF knockout HEK-293T cell lysate (20µg)

Lanes 1-2: Merged signal (red and green). Green - <u>ab231155</u> observed at 23 kDa. Red - loading control <u>ab8245</u> observed at 37 kDa.

ab231155 Anti-Cyclophilin F antibody [EPR11311-121] was shown to specifically react with Cyclophilin F in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266077 (knockout cell lysate ab257039) was used. Wild-type and Cyclophilin F knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab231155 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.

Mut CCGTCCCGCGCTCCGTGCCGCTGCCGCCTCCCCCGCGCCCGCGCCTGCAGCAAGGGCTCC

WT CCGTCCCGCGCTCCGTGCCGCTGCCGCCTCCCC GCGGCCCGCGCCTGCAGCAAGGGCTCC

Sanger Sequencing - Human PPIF knockout

HEK293T cell lysate (ab257039)

Homozygous: 1 bp insertion in exon 1

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