# abcam

# Product datasheet

# Human PARK7 (DJ1) knockout HEK-293T cell lysate ab257016

# 3 Images

Overview

Product name Human PARK7 (DJ1) 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: Insertion of the selection cassette in

exon 2.

Passage number <20

**Knockout validation** Sanger Sequencing, Western Blot (WB)

**Reconstitution notes**To 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 <u>here</u>. 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 -

 $20^{\circ}\text{C}$  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.

It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses.

This product is subject to limited use licenses from The Broad Institute, ERS Genomics Limited and Sigma-Aldrich Co. LLC, and is developed with patented technology. For full details of the

licenses and patents please refer to our limited use license and patent pages.

1

#### **Properties**

#### Storage instructions

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

Components	1 kit
ab260934 - Human PARK7 knockout HEK293T cell lysate	1 x 100µg
ab255553 - Human wild-type HEK293T cell lysate	1 x 100µg

Cell type

epithelial

**STR Analysis** 

 $Amelogenin\,X\,\,D5S818:\,8,\,9\,\,D13S317:\,12,\,14\,\,D7S820:\,11\,\,D16S539:\,9,\,13\,\,vWA:\,16,\,19\,\,TH01:\,12,\,14\,\,D7S820:\,11\,\,D16S539:\,12,\,13\,\,vWA:\,14,\,14\,\,D7S820:\,14\,\,D16S539:\,14,\,14\,\,D16$ 

7, 9.3 TPOX: 11 CSF1PO: 11, 12

#### **Target**

#### **Function**

Protects cells against oxidative stress and cell death. Plays a role in regulating expression or stability of the mitochondrial uncoupling proteins SLC25A14 and SLC25A27 in dopaminergic neurons of the substantia nigra pars compacta and attenuates the oxidative stress induced by calcium entry into the neurons via L-type channels during pacemaking. Eliminates hydrogen peroxide and protects cells against hydrogen peroxide-induced cell death. May act as an atypical peroxiredoxin-like peroxidase that scavenges hydrogen peroxide. Following removal of a Cterminal peptide, displays protease activity and enhanced cytoprotective action against oxidative stress-induced apoptosis. Stabilizes NFE2L2 by preventing its association with KEAP1 and its subsequent ubiquitination. Binds to OTUD7B and inhibits its deubiquitinating activity. Enhances RELA nuclear translocation. Binds to a number of mRNAs containing multiple copies of GG or CC motifs and partially inhibits their translation but dissociates following oxidative stress. Required for correct mitochondrial morphology and function and for autophagy of dysfunctional mitochondria. Regulates astrocyte inflammatory responses. Acts as a positive regulator of androgen receptordependent transcription. Prevents aggregation of SNCA. Plays a role in fertilization. Has no proteolytic activity. Has cell-growth promoting activity and transforming activity. May function as a redox-sensitive chaperone.

## Tissue specificity

Highly expressed in pancreas, kidney, skeletal muscle, liver, testis and heart. Detected at slightly lower levels in placenta and brain. Detected in astrocytes, Sertoli cells, spermatogonia, spermatids and spermatozoa.

#### Involvement in disease

Defects in PARK7 are the cause of Parkinson disease type 7 (PARK7) [MIM:606324]. A neurodegenerative disorder characterized by resting tremor, postural tremor, bradykinesia, muscular rigidity, anxiety and psychotic episodes. PARK7 has onset before 40 years, slow progression and initial good response to levodopa. Some patients may show traits reminiscent of amyotrophic lateral sclerosis-parkinsonism/dementia complex (Guam disease).

## Sequence similarities

Belongs to the peptidase C56 family.

# Post-translational modifications

Sumoylated on Lys-130 by PIAS2 or PIAS4; which is enhanced after ultraviolet irradiation and essential for cell-growth promoting activity and transforming activity.

Cys-106 is easily oxidized to sulfinic acid.

Undergoes cleavage of a C-terminal peptide and subsequent activation of protease activity in response to oxidative stress.

#### **Cellular localization**

Cytoplasm. Nucleus. Mitochondrion. Under normal conditions, located predominantly in the cytoplasm and, to a lesser extent, in the nucleus and mitochondrion. Translocates to the mitochondrion and subsequently to the nucleus in response to oxidative stress and exerts an increased cytoprotective effect against oxidative damage. Detected in tau inclusions in brains from neurodegenerative disease patients.

#### **Applications**

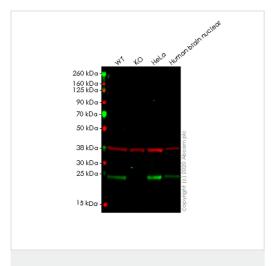
#### The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab257016 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: 20 kDa.

#### **Images**



Western blot - Human PARK7 (DJ1) knockout HEK293T cell lysate (ab257016)

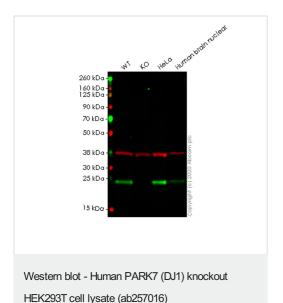
Lane 1: Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate (20 ug)

Lane 2: PARK7 knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate (20 ug)

Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate (20 ug)

Lane 4: Human brain nuclear fraction tissue lysate (20 ug)

<u>ab76241</u> was shown to specifically react with PARK7/DJ1 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line <u>ab266338</u> (knockout cell lysate ab257016) was used. Wild-type and PARK7/DJ1 knockout samples were subjected to SDS-PAGE. <u>ab76241</u> and Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



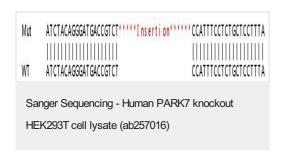
Lane 1:Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate (20 ug)

Lane 2: PARK7 knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate (20 ug)

Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate (20 ug)

Lane 4: Human brain nuclear fraction tissue lysate (20 ug)

ab76008 was shown to specifically react with PARK7/DJ1 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab266338 (knockout cell lysate ab257016) was used. Wild-type and PARK7/DJ1 knockout samples were subjected to SDS-PAGE. ab76008 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.



Homozygous: Insertion of the selection cassette in exon 2

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

#### Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- · Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you

• We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <a href="https://www.abcam.com/abpromise">https://www.abcam.com/abpromise</a> or contact our technical team.

## Terms and conditions

• Guarantee only valid for products bought direct from Abcam or one of our authorized distributors