

Product datasheet

Human PARK7 (DJ1) knockout HEK293T cell line  
ab266338

4 Images

Overview

<b>Product name</b>	Human PARK7 (DJ1) knockout HEK293T cell line
<b>Parental Cell Line</b>	HEK293T
<b>Organism</b>	Human
<b>Mutation description</b>	Knockout achieved by using CRISPR/Cas9, Homozygous: Insertion of the selection cassette in exon 2
<b>Passage number</b>	<20
<b>Knockout validation</b>	Sanger Sequencing, Western Blot (WB)
<b>Tested applications</b>	<b>Suitable for:</b> WB
<b>Biosafety level</b>	2
<b>General notes</b>	<p><b>Recommended control:</b> Human wild-type HEK293T cell line (<a href="#">ab255449</a>). 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.</p> <p><b>Cryopreservation cell medium:</b> Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p><b>Culture medium:</b> DMEM (High Glucose) + 10% FBS</p> <p><b>Initial handling guidelines:</b> 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.</p> <ol style="list-style-type: none"> <li>1. Thaw the vial in 37°C water bath approximately 1-2 minutes.</li> <li>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 <b>culture medium</b>, wash vial with an additional 0.8 ml <b>culture medium</b> (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.</li> <li>3. Resuspend the cell pellet in 5 ml pre-warmed <b>culture medium</b> and count using a haemocytometer (<a href="#">Click here to view haemocytometer protocol</a>) or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of <math>2 \times 10^4</math> cells/cm<sup>2</sup>. This should allow for confluency within 48 hours. Seeding density is given as a guide only and should be scaled to align with individual lab schedules.</li> <li>4. Incubate the culture at 37°C incubator with 5% CO<sub>2</sub>. Cultures should be monitored daily.</li> </ol> <p><b>Subculture guidelines:</b></p>

All seeding densities should be based on cell counts gained by established methods. A guide seeding density of  $2 \times 10^4$  cells/cm<sup>2</sup> is recommended for confluency (80-90% confluence) within 48 hours. 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. [Click here to view the Mammalian cell tissue culture protocol](#)

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## Properties

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<b>Number of cells</b>	1 x 10 <sup>6</sup> cells/vial, 1 mL
<b>Viability</b>	~90%
<b>Adherent /Suspension</b>	Adherent
<b>Tissue</b>	Kidney
<b>Cell type</b>	epithelial
<b>STR Analysis</b>	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
<b>Mycoplasma free</b>	Yes
<b>Storage instructions</b>	Shipped on Dry Ice. Store in liquid nitrogen.
<b>Storage buffer</b>	Constituents: 8.7% DMSO, 2% Cellulose, methyl ether

## Target

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<b>Function</b>	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 C-terminal 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 receptor-dependent 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.
<b>Tissue specificity</b>	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.
<b>Involvement in disease</b>	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.

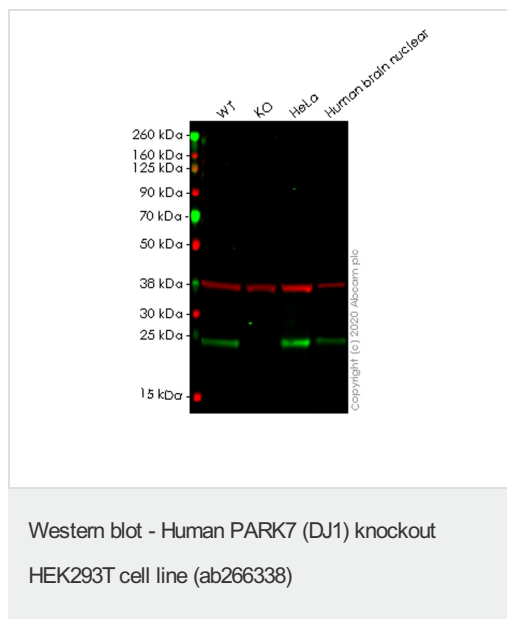
### Applications

Our [Abpromise guarantee](#) covers the use of **ab266338** 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



**All lanes** : Anti-PARK7/DJ1 antibody [EP2816Y] ([ab76241](#)) at 1/1000 dilution

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

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

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

**Lane 4** : Human brain nuclear fraction tissue 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

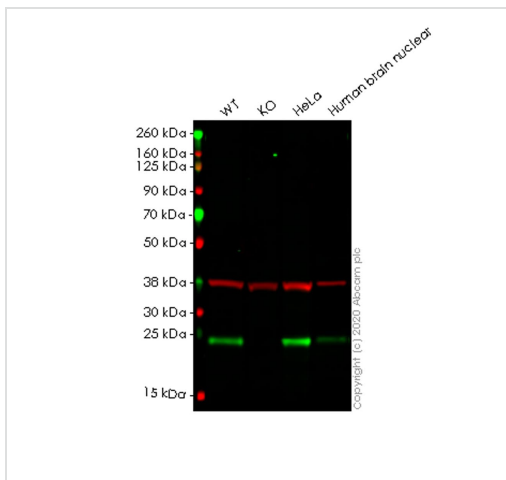
**Predicted band size:** 20 kDa

**Observed band size:** 24 kDa

[why is the actual band size different from the predicted?](#)

**Lanes 1-4:** Merged signal (red and green). Green - [ab76241](#) observed at 24 kDa. Red - loading control [ab8245](#) observed at 36 kDa.

[ab76241](#) Anti-PARK7/DJ1 antibody [EP2816Y] 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. [ab76241](#) 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 PARK7 (DJ1) knockout HEK293T cell line ([ab266338](#))

**All lanes :** Anti-PARK7/DJ1 antibody [EP2815Y] ([ab76008](#)) at 1/1000 dilution

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

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

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

**Lane 4 :** Human brain nuclear fraction tissue 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:** 20 kDa

**Observed band size:** 24 kDa [why is the actual band size different from the predicted?](#)

**Lanes 1-4:** Merged signal (red and green). Green - [ab76008](#) observed at 24 kDa. Red - loading control [ab8245](#) observed at 36 kDa.

[ab76008](#) Anti-PARK7/DJ1 antibody [EP2815Y] 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.

Mut	ATCTACAGGGATGACCGTCT*****Insertion*****CCATTTCTCTGCTCCTTTA
WT	ATCTACAGGGATGACCGTCTCCATTTCTCTGCTCCTTTA

Sanger Sequencing - Human PARK7 knockout  
HEK293T cell line (ab266338)

Homozygous: Insertion of the selection cassette in exon 2

Human PARK7 (DJ1) knockout HEK293T cell line  
(ab266338)

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

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