abcam

Product datasheet

Human NDUFS3 knockout HEK-293T cell lysate ab257556

5 Images

Overview

Product name	Human NDUFS3 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: 19 bp deletion in exon 1.
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 **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. 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 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**.
Tested applications	**Suitable for:** WB

Properties

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

Components	1 kit
ab260280 - Human NDUFS3 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 Core subunit of the mitochondrial membrane respiratory chain NADH dehydrogenase (Complex I) that is believed to belong to the minimal assembly required for catalysis. Complex I functions in the transfer of electrons from NADH to the respiratory chain. The immediate electron acceptor for the enzyme is believed to be ubiquinone.

Sequence similarities Belongs to the complex I 30 kDa subunit family.

Cellular localization Mitochondrion inner membrane.

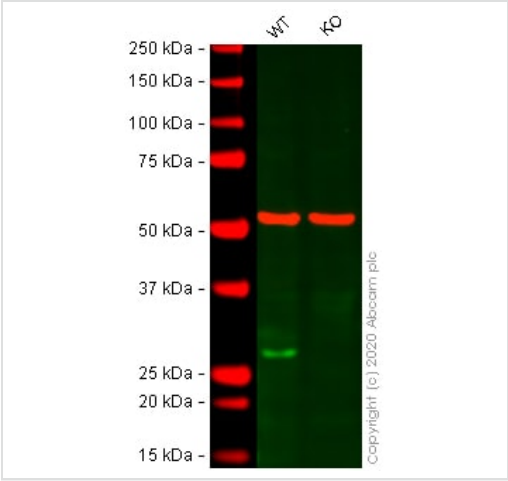
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab257556 in the following tested applications.

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

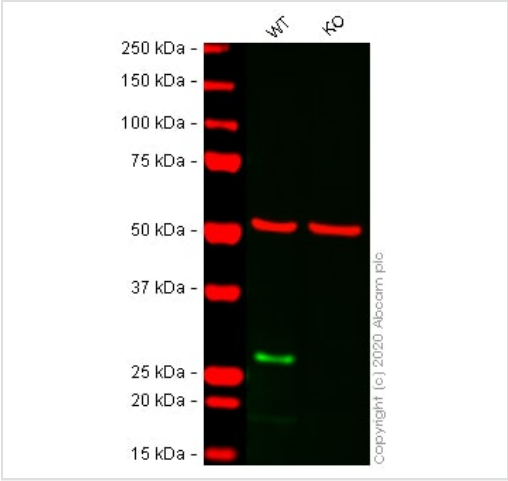
Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 30 kDa.

Images



Western blot - Human NDUFS3 knockout HEK293T cell lysate (ab257556)

Lane 1: Wild-type HEK-293T cell lysate 20 ug
Lane 2: NDUFS3 knockout HEK-293T cell lysate 20 ug
Lanes 1 - 2: Merged signal (red and green). Green - **ab177471** observed at 27 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.
ab177471 was shown to react with NDUFS3 in wild-type HEK-293T cells in western blot with loss of signal observed in NDUFS3 knockout cell line **ab266419** (NDUFS3 knockout cell lysate ab257556). Wild-type and NDUFS3 knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab177471** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively.. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



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Lane 1: Wild-type HEK-293T cell lysate 20 ug
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Lanes 1 - 2: Merged signal (red and green). Green - **ab183733** observed at 27 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.
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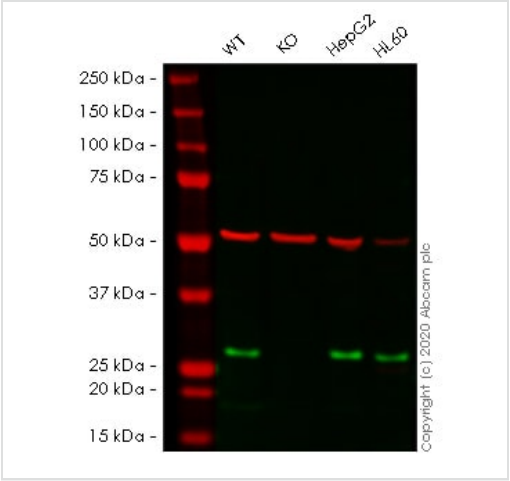


Western blot - Human NDUFS3 knockout HEK293T cell lysate (ab257556)

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

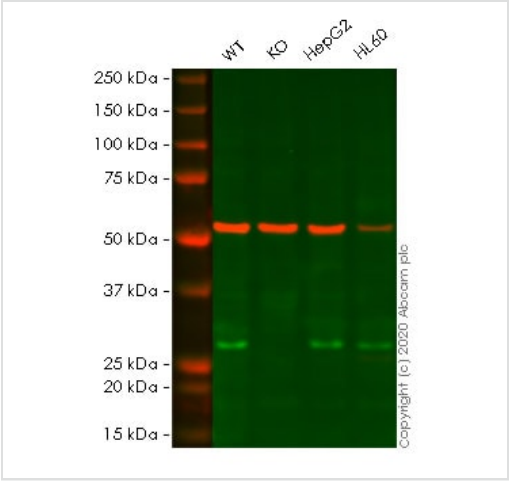
Lane 2: NDUFS3 knockout HEK-293T cell lysate (20µg)

Lane 3: HepG2 cell lysate (20µg)

Lane 4: HL60 cell lysate (20µg)

Lanes 1- 4: Merged signal (red and green). Green - **ab183733** observed at 30 kDa. Red - loading control **ab7291** observed at 50 kDa.

ab183733 Rabbit monoclonal [EPR12781] to NDUFS3 was shown to specifically react with NDUFS3 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line **ab266419** (knockout cell lysate ab257556) was used. Wild-type and NDUFS3 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. **ab183733** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) were incubated overnight at 4°C at 1 in 10000 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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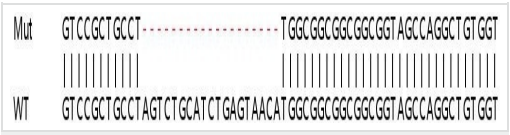
Lane 4: HL60 cell lysate (20µg)

Lanes 1- 4: Merged signal (red and green). Green - **ab177471** observed at 30 kDa. Red - loading control **ab7291** observed at 50 kDa.

ab177471 Rabbit monoclonal [EPR12782] to NDUFS3 - C-terminal was shown to specifically react with NDUFS3 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line **ab266419** (knockout cell lysate ab257556) was

used. Wild-type and NDUFS3 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. **ab177471** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) 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.



Sanger Sequencing - Human NDUFS3 knockout HEK293T cell lysate (ab257556)

Homozygous: 19 bp deletion in exon 1

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