abcam

Product datasheet

Human HDAC2 knockout HEK-293T cell lysate ab256938

4 Images

Overview

Product name	Human HDAC2 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, 1 bp insertion in exon2 and 41 bp insertion in exon2.
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, 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**.
Tested applications	**Suitable for:** WB

Properties

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

Components	1 kit
ab260186 - Human HDAC2 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 Responsible for the deacetylation of lysine residues on the N-terminal part of the core histones (H2A, H2B, H3 and H4). Histone deacetylation gives a tag for epigenetic repression and plays an important role in transcriptional regulation, cell cycle progression and developmental events. Histone deacetylases act via the formation of large multiprotein complexes.
Forms transcriptional repressor complexes by associating with MAD, SIN3, YY1 and N-COR. Interacts in the late S-phase of DNA-replication with DNMT1 in the other transcriptional repressor complex composed of DNMT1, DMAP1, PCNA, CAF1. Deacetylates TSHZ3 and regulates its transcriptional repressor activity.

Tissue specificity Widely expressed; lower levels in brain and lung.

Sequence similarities Belongs to the histone deacetylase family. HD type 1 subfamily.

Post-translational modifications S-nitrosylated by GAPDH. In neurons, S-Nitrosylation at Cys-262 and Cys-274 does not affect the enzyme activity but abolishes chromatin-binding, leading to increases acetylation of histones and activate genes that are associated with neuronal development. In embryonic cortical neurons, S-Nitrosylation regulates dendritic growth and branching.

Cellular localization Nucleus.

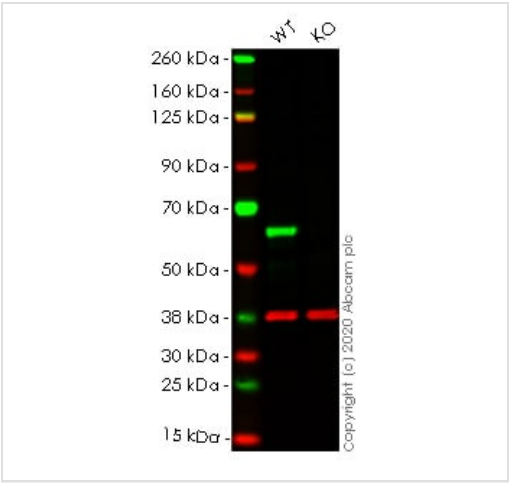
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab256938 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: 55 kDa.

Images



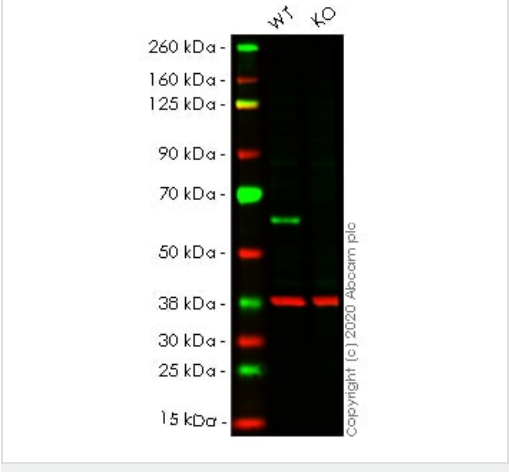
Western blot - Human HDAC2 knockout HEK293T cell lysate (ab256938)

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

Lane 2: HDAC2 knockout HEK-293T cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - **ab219053** observed at 55 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab219053 Rabbit monoclonal [EPR20117] to HDAC2 was shown to specifically react with HDAC2 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line **ab266589** (knockout cell lysate ab256938) was used. Wild-type and HDAC2 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. **ab219053** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4˚C at 1 in 1000 µg/ml 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 HDAC2 knockout HEK293T cell lysate (ab256938)

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

Lane 2: HDAC2 knockout HEK-293T cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - **ab124974** observed at 55 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab124974 Rabbit monoclonal [EPR5001] to HDAC2 - ChIP Grade was shown to specifically react with HDAC2 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line **ab266589** (knockout cell lysate ab256938) was used. Wild-type and HDAC2 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. **ab124974** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) 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.

Sanger Sequencing - Human HDAC2 knockout HEK293T cell lysate (ab256938)

Allele-1: 1 bp insertion in exon2

Sanger Sequencing - Human HDAC2 knockout HEK293T cell lysate (ab256938)

Allele-2: 41 bp insertion in exon2

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