abcam

Product datasheet

Human CELF1 (CUG-BP1) knockout HEK-293T cell lysate ab257390

4 Images

Overview

Product name Human CELF1 (CUG-BP1) knockout HEK-293T cell lysate

Product overview

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line HEK293T Human

Organism

Mutation description Knockout achieved by using CRISPR/Cas9, 4 bp deletion in exon 2 and 5 bp deletion 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 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
ab260979 - Human CELF1 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

RNA-binding protein implicated in the regulation of several post-transcriptional events. Involved in pre-mRNA alternative splicing, mRNA translation and stability. Mediates exon inclusion and/or exclusion in pre-mRNA that are subject to tissue-specific and developmentally regulated alternative splicing. Specifically activates exon 5 inclusion of cardiac isoforms of TNNT2 during heart remodeling at the juvenile to adult transition. Acts as both an activator and repressor of a pair of coregulated exons: promotes inclusion of the smooth muscle (SM) exon but exclusion of the non-muscle (NM) exon in actinin pre-mRNAs. Activates SM exon 5 inclusion by antagonizing the repressive effect of PTB. Promotes exclusion of exon 11 of the INSR pre-mRNA. Inhibits, together with HNRNPH1, insulin receptor (IR) pre-mRNA exon 11 inclusion in myoblast. Increases translation and controls the choice of translation initiation codon of CEBPB mRNA. Increases mRNA translation of CEBPB in aging liver (By similarity). Increases translation of CDKN1A mRNA by antagonizing the repressive effect of CALR3. Mediates rapid cytoplasmic mRNA deadenylation. Recruits the deadenylase PARN to the poly(A) tail of EDEN-containing mRNAs to promote their deadenylation. Required for completion of spermatogenesis (By similarity). Binds to (CUG)n triplet repeats in the 3'-UTR of transcripts such as DMPK and to Bruno response elements (BREs). Binds to muscle-specific splicing enhancer (MSE) intronic sites flanking the alternative exon 5 of TNNT2 pre-mRNA. Binds to AU-rich sequences (AREs or EDEN-like) localized in the 3'-UTR of JUN and FOS mRNAs. Binds to the IR RNA. Binds to the 5'-region of CDKN1A and CEBPB mRNAs. Binds with the 5'-region of CEBPB mRNA in aging liver.

Tissue specificity

Ubiquitous.

Sequence similarities

Belongs to the CELF/BRUNOL family.

Contains 3 RRM (RNA recognition motif) domains.

Post-translational

modifications

Phosphorylated. Its phosphorylation status increases in senescent cells.

Cellular localization

Nucleus. Cytoplasm. RNA-binding activity is detected in both nuclear and cytoplasmic

compartments.

Applications

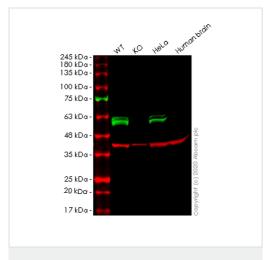
The Abpromise guarantee

Our **Abpromise quarantee** covers the use of ab257390 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: 52 kDa.

Images



Western blot - Human CELF1 knockout HEK293T cell lysate (ab257390)

Lane 1: Wild-type HEK293T cell lysate (20 µg)

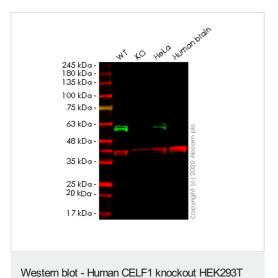
Lane 2: CELF1 knockout HEK293T cell lysate (20 µg)

Lane 3: HeLa cell lysate (20 µg)

Lane 4: Human brain tissue lysate (20 µg)

Lanes 1-4: Merged signal (red and green). Green - <u>ab129115</u> observed at 52 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

ab129115 Anti-CUG-BP1 antibody [EPR8298(B)] was shown to specifically react with CUG-BP1 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266086 (knockout cell lysate ab257390) was used. Wild-type and CUG-BP1 knockout samples were subjected to SDS-PAGE. ab129115 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 lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



cell lysate (ab257390)

Lane 1: Wild-type HEK293T cell lysate (20 µg)

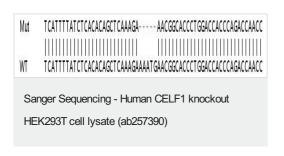
Lane 2: CELF1 knockout HEK293T cell lysate (20 µg)

Lane 3: HeLa cell lysate (20 µg)

Lane 4: Human brain tissue lysate (20 µg)

Lanes 1-4: Merged signal (red and green). Green - <u>ab9549</u> observed at 52 kDa. Red - loading control <u>ab181602</u> observed at 36 kDa.

ab9549 Anti-CUG-BP1 antibody [3B1] was shown to specifically react with CUG-BP1 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266086 (knockout cell lysate ab257390) was used. Wild-type and CUG-BP1 knockout samples were subjected to SDS-PAGE. ab9549 and Anti-GAPDH antibody[EPR16891] - Loading Control (ab181602) 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® 680RD) preadsorbed (ab216777) and Goat anti-Mouse lgG H&L (IRDye® 800CW) preadsorbed (ab216772) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Allele-1: 5 bp deletion in exon 2

Mut	TCATTTTATCTCACACAGCTCAAAGAGAACGGCACCCTGGACCACCCAGACCAACC		
WT	TCATTTTATCTCACACAGCTCAAAGAAAATGAACGGCACCCTGGACCACCCAGACCAACC		
Sanger Sequencing - Human CELF1 knockout			
Sanger Sequencing - Human CELFT Knockout			
HEK293T cell lysate (ab257390)			

Allele-2: 4 bp deletion in exon 2

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