abcam

Product datasheet

Human TRIP10 (Cip4) knockout HEK-293T cell lysate ab258251

3 Images

Overview

Organism

Product name Human TRIP10 (Cip4) knockout HEK-293T cell lysate

Product overview

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line HEK293T

Mutation description Knockout achieved by using CRISPR/Cas9, 14 bp deletion in exon2.

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

Human

Reconstitution notesTo 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.

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products that contain European Authorisation list (Annex XIV) substances.

It is the responsibility of our customers to check the necessity of application of REACH

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relevant patents please refer to our <u>limited use license</u> and <u>patent pages</u>.

Tested applications Suitable for: WB

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Properties

Storage instructions

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

Components	1 kit
ab262353 - Human TRIP10 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

Required for translocation of GLUT4 to the plasma membrane in response to insulin signaling (By similarity). Required to coordinate membrane tubulation with reorganization of the actin cytoskeleton during endocytosis. Binds to lipids such as phosphatidylinositol 4,5-bisphosphate and phosphatidylserine and promotes membrane invagination and the formation of tubules. Also promotes CDC42-induced actin polymerization by recruiting WASL/N-WASP which in turn activates the Arp2/3 complex. Actin polymerization may promote the fission of membrane tubules to form endocytic vesicles. Required for the formation of podosomes, actin-rich adhesion structures specific to monocyte-derived cells. May be required for the lysosomal retention of FASLG/FASL.

Tissue specificity

Expressed in brain, colon, heart, kidney, liver, lung, megakaryocyte, ovary, pancreas, peripheral blood lymphocytes, placenta, prostate, skeletal muscle, small intestine, spleen, testis, thymus and trachea.

Sequence similarities

Belongs to the FNBP1 family. Contains 1 FCH domain. Contains 1 REM (Hr1) repeat. Contains 1 SH3 domain.

Post-translational modifications

Tyrosine phosphorylated. Also phosphorylated by PKA.

Cellular localization

Cytoplasm > perinuclear region and Cytoplasm > cytoskeleton. Cytoplasm > cell cortex. Lysosome. Golgi apparatus. Cell membrane. Cell projection > phagocytic cup. Translocates to the plasma membrane in response to insulin stimulation, and this may require active RHOQ (By similarity). Localizes to cortical regions coincident with F-actin, to lysosomes and to sites of phagocytosis in macrophages. Also localizes to the Golgi, and this requires AKAP9.

Applications

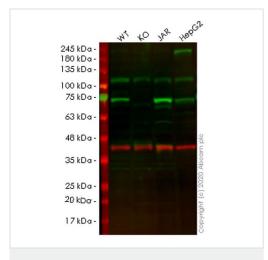
The Abpromise guarantee

Our Abpromise quarantee covers the use of ab258251 in the following tested applications.

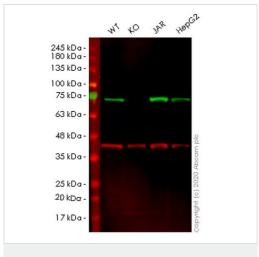
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 68 kDa.

Images



Western blot - Human TRIP10 (Cip4) knockout HEK293T cell lysate (ab258251)



Western blot - Human TRIP10 (Cip4) knockout HEK293T cell lysate (ab258251)

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

Lane 2:TRIP10 knockout HEK293T cell lysate (20 ug)

Lane 3: JAR cell lysate (20 ug)

Lane 4:HepG2 cell lysate (20 ug)

ab108313 was shown to specifically react with Cip4 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266428 (knockout cell lysate ab258251) was used. Wild-type and Cip4 knockout samples were subjected to SDS-PAGE. ab108313 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.

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

Lane 2:TRIP10 knockout HEK293T cell lysate (20 ug)

Lane 3: JAR cell lysate (20 ug)

Lane 4: HepG2 cell lysate (20 ug)

ab108277 was shown to specifically react with Cip4 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266428 (knockout cell lysate ab258251) was used. Wild-type and Cip4 knockout samples were subjected to SDS-PAGE. ab108277 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	GTAGCGTCCACCCCAGGGTTTGGCGTAAGCCTGTTCCACTTCGGTG
	WT	GTAGCGTCCACCCCAGGGTCTCACCGCAGTTGTTTGGCGTAAGCCTGTTCCACTTCGGTG
	Sa	nger Sequencing - Human TRIP10 knockout
HEK293T cell lysate (ab258251)		

Homozygous: 14 bp deletion in exon2

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