abcam

Product datasheet

Human PKN1 knockout HEK-293T cell lysate ab258586

画像数3

製品の概要

製品名	Human PKN1 knockout HEK-293T cell lysate	
製品の概要		
	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 Insertion of the selection cassette in exon2.	
Passage number	<20	
Knockout validation	Sanger Sequencing, Western Blot (WB)	
Reconstitution notes	To use as WB control, resuspend the lyophilizate in 50 μ L 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.	
特記事項	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). <i>This means that the protein of interest is denatured.</i> If you require a native form of the protein please use the live cell version - found <u>here</u> . 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 .	

製品の特性

保存方法

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

内容	1 kit
ab261287 - Human PKN1 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	

ターゲット情報

機能	PKC-related serine/threonine-protein kinase involved in various processes such as regulation of the intermediate filaments of the actin cytoskeleton, cell migration, tumor cell invasion and transcription regulation. Regulates the cytoskeletal network by phosphorylating proteins such as VIM and neurofilament proteins NEFH, NEFL and NEFM, leading to inhibit their polymerization. Phosphorylates 'Ser-575', 'Ser-637' and 'Ser-669' of MAPT/Tau, lowering its ability to bind to microtubules, resulting in disruption of tubulin assembly. Acts as a key coactivator of androgen receptor (ANDR)-dependent transcription, by being recruited to ANDR target genes and specifically mediating phosphorylation of 'Thr-11' of histone H3 (H3T11ph), a specific tag for epigenetic transcriptional activation that promotes demethylation of histone H3 'Lys-9' (H3K9me) by KDM4C/JMJD2C. Phosphorylates HDAC5, HDAC7 and HDAC9, leading to impair their import in the nucleus. Phosphorylates 'Thr-38' of PPP1R14A, 'Ser-159', 'Ser-163' and 'Ser-170' of MARCKS, and GFAP. Able to phosphorylate RPS6 in vitro.	
組織特異性	Found ubiquitously. Expressed in heart, brain, placenta, lung, skeletal muscle, kidney and pancreas. Expressed in numerous tumor cell lines, especially in breast tumor cells.	
配列類似性	Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family. PKC subfamily. Contains 1 AGC-kinase C-terminal domain. Contains 1 C2 domain. Contains 1 protein kinase domain. Contains 3 REM (Hr1) repeats.	
ドメイン	The C1 domain does not bind the diacylglycerol (DAG).	
翻訳後修飾	Autophosphorylated; preferably on serine. Phosphorylated during mitosis. Activated by limited proteolysis with trypsin.	
細胞内局在	Cytoplasm. Nucleus. Endosome. Cell membrane. Cleavage furrow. Midbody. Associates with chromatin in a ligand-dependent manner. Localization to endosomes is mediated via its interaction with RHOB. Association to the cell membrane is dependent on Ser-374 phosphorylation. Accumulates during telophase at the cleavage furrow and finally concentrates around the midbody in cytokinesis.	

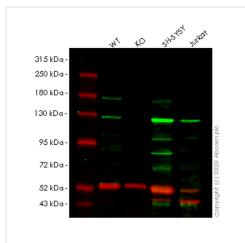
The Abpromise guarantee

Abpromise保証は、次のテスト済みアプリケーションにおけるab258586の使用に適用されます

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 104 kDa.

画像



Western blot - Human PKN1 knockout HEK293T cell lysate (ab258586)

Lane 1: Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate (20 ug)

Lane 2: PKN1 knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate (20 ug)

Lane 3:SH-SY5Y (Human neuroblastoma cell line from bone marrow) whole cell lysate (20 ug)

Lane 4: Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate (20 ug)

ab108976 was shown to specifically react with PKN in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line **ab266599** (knockout cell lysate ab258586) was used. Wildtype and PKN knockout samples were subjected to SDS-PAGE. **ab108976** 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.

Allele-1: 1 bp insertion in exon2

 Mut
 T GGAGC GGGAGC GGCT GC GGC GGGAAAT CCC GC AAGGAGCT GAAGCT GAAGGAGGGT GCT

 WI
 T GGAGC GGGAGC GGCT GC GGC GGGAAAT CC
 GC AAGGAGCT GAAGCT GAAGGAGGT GCT

Sanger Sequencing - Human PKN1 knockout HEK293T cell lysate (ab258586)



Allele-2: Insertion of the selection cassette in exon2

Sanger Sequencing - Human PKN1 knockout HEK293T cell lysate (ab258586)

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