

Product datasheet

Human PHKA1 knockout HEK-293T cell lysate ab258111

2 Images

Overview

Product name	Human PHKA1 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, 2 bp deletion in exon14.
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. <i>*Usage of SDS sample buffer is not recommended with these lyophilized lysates.</i>

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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Properties

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

Components	1 kit
ab262304 - Human PHKA1 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 Phosphorylase b kinase catalyzes the phosphorylation of serine in certain substrates, including troponin I. The alpha chain may bind calmodulin.

Tissue specificity Muscle specific. Isoform 1 is predominant in vastus lateralis muscle. Isoform 2 predominates slightly in heart, and it predominates clearly in the other tissues tested.

Pathway Glycan biosynthesis; glycogen metabolism.

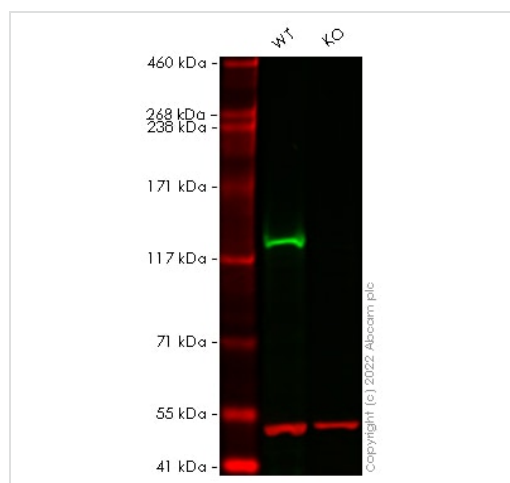
Involvement in disease Glycogen storage disease 9D (GSD9D) [MIM:300559]: A metabolic disorder characterized by slowly progressive, predominantly distal muscle weakness and atrophy. Clinical features include exercise intolerance with early fatigability, pain, cramps and occasionally myoglobinuria. Note=The disease is caused by mutations affecting the gene represented in this entry.

Sequence similarities Belongs to the phosphorylase b kinase regulatory chain family.

Post-translational modifications Although the final Cys may be farnesylated, the terminal tripeptide is probably not removed, and the C-terminus is not methylated.

Cellular localization Cell membrane.

Images



Western Blot Human PHKA1 KO HEK283T cell lysate

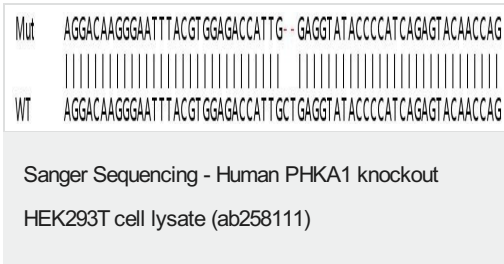
False colour image of Western blot: Anti-PHKA1 antibody [EPR12118] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] ([ab7291](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, [ab176338](#) was shown to bind specifically to PHKA1. A band was observed at 130 kDa in wild-type HEK-293T cell lysates with no signal observed at this size in PHKA1 knockout cell line [ab267337](#) (knockout cell lysate ab258111).

To generate this image, wild-type and PHKA1 knockout HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 5 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C.

Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

Lane 1: Wild-type HEK-293T cell lysate, 20ug

Lane 2: PHKA1 knockout HEK-293T cell lysate, 20ug



Homozygous: 2 bp deletion in exon14

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