abcam

Product datasheet

Human TMUB1 knockout HeLa cell lysate ab258237

3 Images

Overview

Product name	Human TMUB1 knockout HeLa cell lysate
Product overview	Knockout cell lysate achieved by CRISPR/Cas9.
	Knockout profile: Only the long form of the protein (IHOPS, 27 kDa) has been knocked-out from the parental cell line. The band observed in the KO lysate at 21 kDa is likely to represent a short form of the protein (sHOPS) (doi: 10.4161/cc.27054). We have not investigated the function of the remaining form of the protein.
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon2 and 1 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 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.
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). <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 .								
Tested applications	Suitable for: WB, Sanger Sequencing								

Properties

Storage instructions	Store at -80°C. Please refer to protocols.								
Components		1 kit							
ab261184 - Human TMUB1 knock	out HeLa cell lysate	1 x 100µg							
ab255929 - Human wild-type HeLa	a cell lysate	1 x 100µg							
Cell type	epithelial								
Disease	se Adenocarcinoma								
Gender	Female								
STR Analysis	Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8,12 CSF1PO: 9, 10								
Target									
Function	May contribute to the regulation of translation during cell-cycle progressio regulation of cell proliferation.	n. May contribute to the							

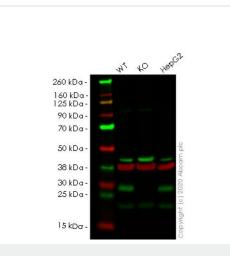
Sequence similaritiesContains 1 ubiquitin-like domain.Cellular localizationMembrane. Cytoplasm. Nucleus. Predominantly nuclear during growth arrest (By similarity).
Actively exported from the nucleus in dividing cells.

Applications

The Abpromise guaranteeOur Abpromise guaranteecovers the use of ab258237 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: 26 kDa.
Sanger Sequencing		Use at an assay dependent concentration.

Images



Western blot - Human TMUB1 knockout HeLa cell lysate (ab258237)

Lane 1: Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate (20µg)

Lane 2: TMUB1 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) cell lysate (20µg)

Lane 3: HepG2 (Human liver hepatocellular carcinoma cell line) cell lysate cell lysate (20µg)

Lanes 1-3: Merged signal (red and green). Green - <u>ab180586</u> observed at 27 kDa. Red - loading control <u>ab8245</u> observed at 37 kDa.

ab180586 Recombinant Anti-TMUB1 antibody [EPR14066] was shown to specifically react with TMUB1 in wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) cells in western blot. Loss of signal was observed when knockout cell line **ab265852** (knockout cell lysate ab258237) was used. Wild-type and TMUB1 knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% nonfat dried milk. **ab180586** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 10000 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 deletion in exon2

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				111		111											$\ $		
TGAAGGG																			

cell lysate (ab258237)

Sanger Sequencing - Human TMUB1 knockout HeLa cell lysate (ab258237)

Allele-2: 1 bp insertion in exon2

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