abcam

Product datasheet

Human DAB2 knockout HeLa cell line ab265731

2 图像

概述

产品名称	人DAB2 knockout HeLa cell line
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 1 bp insertion in exon 3
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
经测试应用	**适用于:** WB
Biosafety level	2
常规说明	**Recommended control:** Human wild-type HeLa cell line (**ab255928**). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1. **Cryopreservation cell medium:** Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose. **Culture medium:** DMEM (High Glucose) + 10% FBS **Initial handling guidelines:** Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability. 1. Thaw the vial in 37°C water bath for approximately 1-2 minutes. 2. Transfer the cell suspension (0.8 mL) to a 15 mL/50 mL conical sterile polypropylene centrifuge tube containing 8.4 mL pre-warmed culture medium, wash vial with an additional 0.8 mL culture medium (total volume 10 mL) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 mL represents minimum recommended dilution. 20 mL represents maximum recommended dilution. 3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of $2x10^4$ cells/cm^2. Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37°C incubator with 5% CO_2. Cultures should be monitored daily. **Subculture guidelines:** All seeding densities should be based on cell counts gained by established methods. A guide seeding density of $2x10^4$ cells/cm^2 is recommended. A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required.

Cells should be passaged when they have achieved 80-90% confluence.
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We will provide viable cells that proliferate on revival.

性能

Number of cells	1 x 10^6 cells/vial, 1 mL
Adherent /Suspension	Adherent
Tissue	Cervix
Cell type	epithelial
Disease	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
Mycoplasma free	Yes
存放说明	Shipped on Dry Ice. Store in liquid nitrogen.
存储溶液	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

靶标

功能	Adapter protein that functions as clathrin-associated sorting protein (CLASP) required for clathrin-mediated endocytosis of selected cargo proteins. Can bind and assemble clathrin, and binds simultaneously to phosphatidylinositol 4,5-bisphosphate (PtdIns(4,5)P2) and cargos containg non-phosphorylated NPXY internalization motifs, such as the LDL receptor, to recruit them to clathrin-coated pits. Can function in clathrin-mediated endocytosis independently of the AP-2 complex. Involved in endocytosis of integrin beta-1; this function seems to redundant with the AP-2 complex and seems to require DAB2 binding to endocytosis accessory EH domain-containing proteins such as EPS15, EPS15L1 and ITSN1. Involved in endocytosis of cystic fibrosis transmembrane conductance regulator/CFTR. Involved in endocytosis of megalin/LRP2 lipoprotein receptor during embryonal development. Required for recycling of the TGF-beta receptor. Involved in CFTR trafficking to the late endosome. Involved in several receptor-mediated signaling pathways. Involved in TGF-beta receptor signaling and facilitates phosphorylation of the signal transducer SMAD2. Mediates TFG-beta-stimulated JNK activation. May inhibit the canoniocal Wnt/beta-catenin signaling pathway by stabilizing the beta-catenin destruction complex through a competing association with axin preventing its dephosphorylation through protein phosphatase 1 (PP1). Sequesters LRP6 towards clathrin-mediated endocytosis, leading to inhibition of Wnt/beta-catenin signaling. May activate non-canonical Wnt signaling. In cell surface growth factor/Ras signaling pathways proposed to inhibit ERK activation by interrupting the binding of GRB2 to SOS1 and to inhibit SRC by preventing its activating phosphorylation at 'Tyr-419'. Proposed to be involved in modulation of androgen receptor (AR) signaling mediated by SRC activation; seems to compete with AR for interaction with SRC. Plays a role in the CSF-1 signal transduction pathway. Plays a role in cellular differentiation. Involved in cell positioning and formation of visceral endoderm (VE) during embryogenesis and proposed to be required in the VE to respond to Nodal signaling coming from the epiblast. Required for the epithelial to

	mesenchymal transition, a process necessary for proper embryonic development. May be involved in myeloid cell differentiation and can induce macrophage adhesion and spreading. May act as a tumor suppressor.
组织特异性	Expressed in deep invaginations, inclusion cysts and the surface epithelial cells of the ovary. Also expressed in breast epithelial cells, spleen, thymus, prostate, testis, macrophages, fibroblasts, lung epithelial cells, placenta, brain stem, heart and small intestine. Expressed in kidney proximal tubular epithelial cells (at protein level).
序列相似性	Contains 1 PID domain.
结构域	The PID domain binds to predominantly non-phosphorylated NPXY internalization motifs present in members of the LDLR and APP family; it also mediates simultaneous binding to phosphatidylinositol 4,5-bisphosphate. The Asn-Pro-Phe (NPF) motifs, which are found in proteins involved in the endocytic pathway, mediate the interaction with the EH domain of EPS15, EPS15R and ITSN1.
翻译后修饰	Phosphorylated. Phosphorylation during mitosis is leading to membrane displacement.
细胞定位	Cytoplasm. Cytoplasmic vesicle, clathrin-coated vesicle membrane. Membrane, clathrin-coated pit. Colocalizes with large insert-containing isoforms of MYO6 at clathrin-coated pits/vesicles. During mitosis is progressively displaced from the membrane and translocated to the cytoplasm.

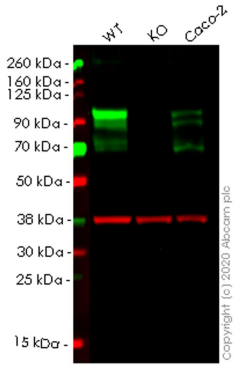
应用

The Abpromise guarantee **Abpromise™承诺保证使用ab265731于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration. Predicted molecular weight: 82 kDa.

图片



Western blot - Human DAB2 knockout HeLa cell line (ab265731)

All lanes : Anti-DAB2 antibody [EPR22881-69] (**ab256524**) at 1/1000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : DAB2 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : Caco-2 (Human colorectal adenocarcinoma cell line) whole cell lysate

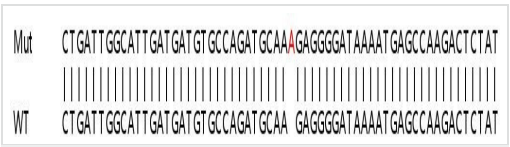
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

Predicted band size: 82 kDa
Observed band size: 95 kDa

Lanes 1-3: Merged signal (red and green). Green - **ab256524** observed at 95 kDa. Red - loading control **ab8245** observed at 36 kDa.

ab256524 Anti-DAB2 antibody [EPR22881-69] was shown to specifically react with DAB2 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265731 (knockout cell lysate **ab257910**) was used. Wild-type and DAB2 knockout samples were subjected to SDS-PAGE. **ab256524** 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.



Sanger Sequencing - Human DAB2 knockout HeLa cell line (ab265731)

Homozygous: 1 bp insertion in exon 3.

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