abcam

Product datasheet

Human FLOT2 (Flotillin 2/ESA) knockout HEK-293T cell line ab266847

3 图像

概述

Parental Cell Line HEK293T
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, Homozygous: 5 bp deletion in exon 4

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

2

经测试应用 适用于: WB

Biosafety level

常规说明

Recommended control: Human wild-type HEK293T cell line (<u>ab255449</u>). 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 2x10⁴ cells/cm². 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% CO₂. 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/cm² is recommended.

1

A partial media change 24 hours prior to subculture may be helpful to encourage growth, if

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 10⁶ cells/vial, 1 mL

Adherent /Suspension Adherent
Tissue Kidney
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

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 May act as a scaffolding protein within caveolar membranes, functionally participating in formation

of caveolae or caveolae-like vesicles. May be involved in epidermal cell adhesion and epidermal $\,$

structure and function.

组织特异性 In skin, expressed in epidermis and epidermal appendages but not in dermis. Expressed in all

layers of the epidermis except the basal layer. In hair follicles, expressed in the suprabasal layer but not the basal layer. Also expressed in melanoma and carcinoma cell lines, fibroblasts and

foreskin melanocytes.

序列相似性 Belongs to the band 7/mec-2 family. Flotillin subfamily.

细胞定位 Cell membrane. Membrane > caveola. Endosome. Membrane-associated protein of caveolae.

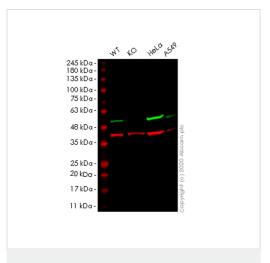
应用

The Abpromise quarantee Abpromise™承诺保证使用ab266847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: 47 kDa.

图片



Western blot - Human FLOT2 knockout HEK293T cell line (ab266847)

All lanes : Anti-Flotillin 2/ESA antibody [EPR14128(B)] (ab181988) at 1/1000 dilution

Lane 1: Wild-type HEK293T cell lysate

Lane 2: FLOT2 knockout HEK293T cell lysate

Lane 3 : HeLa cell lysate Lane 4 : A549 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

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

Predicted band size: 47 kDa **Observed band size:** 47 kDa

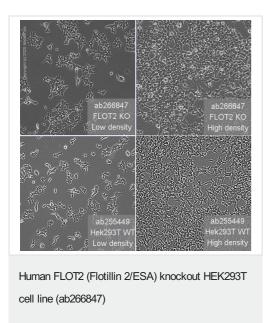
Lanes 1-4: Merged signal (red and green). Green - <u>ab181988</u> observed at 47 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

ab181988 Anti-Flotillin 2/ESA antibody [EPR14128(B)] was shown to specifically react with Flotillin 2/ESA in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266847 (knockout cell lysate ab257442) was used. Wild-type and Flotillin 2/ESA knockout samples were subjected to SDS-PAGE. ab181988 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated at room temperature for 2.5 hours at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Mut CATGACGGAGAAGGAACTCCTGGCCGTG-----TGAGCAGTTTCTGGGTAAGAATGTGCA

WT CATGACGGAGAAGGAACTCCTGGCCGTGGCTTGTGAGCAGTTTCTGGGTAAGAATGTGCA

Sanger Sequencing - Human FLOT2 knockout HEK293T cell line (ab266847) Homozygous: 5 bp deletion in exon4



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