abcam

Product datasheet

Human PPP3CA (Calcineurin A) knockout HeLa cell line ab265130

4 图**像**

概述

产 品名称	人PPP3CA (Calcineurin A) knockout HeLa cell line	
Parental Cell Line	HeLa	
Organism	Human	
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 1 bp deletion in exon 1	
Passage number	<20	
Knockout validation	Sequencing, Western Blot (WB) : 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 (<u>ab255928</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.	
	 Thaw the vial in 37°C water bath for approximately 1-2 minutes. 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. 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. 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.

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.

This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our **limited use license** and **patent pages**.

We will provide viable cells that proliferate on revival.

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	
Antibiotic resistance	Puromycin 1.00µg/ml	
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	Calcium-dependent, calmodulin-stimulated protein phosphatase. This subunit may have a role in	
	the calmodulin activation of calcineurin. Dephosphorylates DNM1L, HSPB1 and SSH1.	
序列相似性	Belongs to the PPP phosphatase family. PP-2B subfamily.	
细 胞定位	Nucleus. Colocalizes with ACTN1 and MYOZ2 at the Z line in heart and skeletal muscle.	

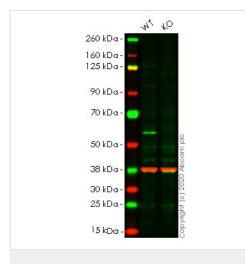
应用

性能

The Abpromise guarantee Abpromise ™</u>承诺保证使用ab265130于以下的经测试应用 "应用说明"如人工具一的识出推荐的扫描系统中,应应具体的系统中应也在用者协定

"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: 59 kDa.



Western blot - Human PPP3CA (Calcineurin A) knockout HeLa cell line (ab265130) All lanes : Anti-Calcineurin A antibody [EPR1670(2)] (<u>ab109412</u>) at 1/10000 dilution

Lane 1 : Wild-type HeLa cell lysate Lane 2 : PPP3CA knockout HeLa cell lysate

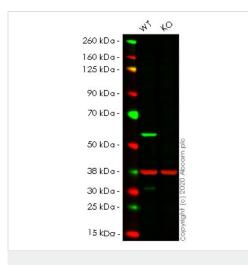
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 59 kDa Observed band size: 59 kDa

Lanes 1-2: Merged signal (red and green). Green - <u>ab109412</u> observed at 59 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) observed at 37 kDa.

ab109412 was shown to react with Calcineurin A in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265130 (knockout cell lysate **ab257181**) was used. Wild-type HeLa and PPP3CA knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab109412** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 10000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye[®]680RD) 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.



Western blot - Human PPP3CA (Calcineurin A) knockout HeLa cell line (ab265130) All lanes : Anti-Calcineurin A antibody [EP1669Y] (<u>ab52761</u>) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate Lane 2 : PPP3CA knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

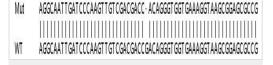
Performed under reducing conditions.

Predicted band size: 59 kDa Observed band size: 59 kDa

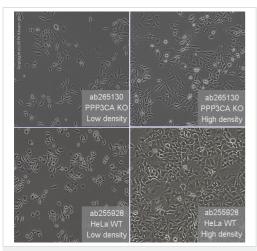
Lanes 1-2: Merged signal (red and green). Green - <u>ab52761</u> observed at 59 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) observed at 37 kDa.

ab52761 was shown to react with Calcineurin A in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265130 (knockout cell lysate **ab257181**) was used. Wild-type HeLa and PPP3CA knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab52761** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye[®]680RD) 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.

Homozygous: 1 bp deletion in exon 1.



Sanger Sequencing - Human PPP3CA knockout HeLa cell line (ab265130)



Cell Culture - Human PPP3CA (Calcineurin A) knockout HeLa cell line (ab265130)

Representative images of PPP3CA knockout HeLa cells, low and high confluency examples (top left and right respectively) and wildtype HeLa cells, low and high confluency (bottom left and right respectively) showing typical adherent, epithelial-like morphology. Images were captured at 10X magnification using a EVOS XL Core microscope.

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