abcam

Product datasheet

Human CHEK2 (Chk2) knockout HeLa cell line ab264815

4 图像

概述

Parental Cell Line HeLa
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, Homozygous: 1 bp insertion in exon 5

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

经测试应用 适用于: WB

Biosafety level

常规说明 Recommended control: Human wild-type HeLa cell line (<u>ab255448</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.

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

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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 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 Regulates cell cycle checkpoints and apoptosis in response to DNA damage, particularly to DNA

double-strand breaks. Inhibits CDC25C phosphatase by phosphorylation on 'Ser-216', preventing the entry into mitosis. May also play a role in meiosis. Regulates the TP53 tumor suppressor

through phosphorylation at 'Thr-18' and 'Ser-20'.

组织特异性 High expression is found in testis, spleen, colon and peripheral blood leukocytes. Low expression

is found in other tissues.

疾病相关 Defects in CHEK2 are associated with Li-Fraumeni syndrome 2 (LFS2) [MIM:609265]; a highly

penetrant familial cancer phenotype usually associated with inherited mutations in p53/TP53. Defects in CHEK2 may be a cause of susceptibility to prostate cancer (PC) [MIM:176807]. It is a malignancy originating in tissues of the prostate. Most prostate cancers are adenocarcinomas that develop in the acini of the prostatic ducts. Other rare histopathologic types of prostate cancer that occur in approximately 5% of patients include small cell carcinoma, mucinous carcinoma, prostatic ductal carcinoma, transitional cell carcinoma, squamous cell carcinoma, basal cell carcinoma, adenoid cystic carcinoma (basaloid), signet-ring cell carcinoma and neuroendocrine

carcinoma.

Defects in CHEK2 are found in some patients with osteogenic sarcoma (OSRC) [MIM:259500].

序列相似性 Belongs to the protein kinase superfamily. CAMK Ser/Thr protein kinase family. CHK2 subfamily.

Contains 1 FHA domain.

Contains 1 protein kinase domain.

翻译**后修**饰 Phosphorylated by PLK4.

细胞定位 Nucleus; Nucleus. Isoform 10 is present throughout the cell and Nucleus > PML body. Nucleus >

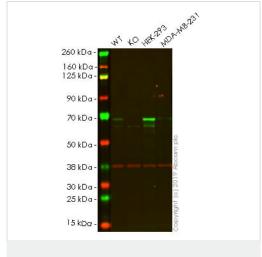
应用

The Abpromise guarantee Abpromise™承诺保证使用ab264815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: 61 kDa.

图片



Western blot - Human CHEK2 (Chk2) knockout HeLa cell line (ab264815) **All lanes :** Anti-Chk2 antibody [EPR19482] (ab207446) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: CHEK2 knockout HeLa cell lysate

Lane 3: HEK-293 cell lysate

Lane 4: MDA-MB-231 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/20000 dilution

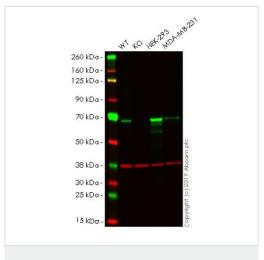
Performed under reducing conditions.

Predicted band size: 61 kDa **Observed band size:** 68 kDa

Lanes 1-4: Merged signal (red and green). Green - <u>ab207446</u> observed at 68 kDa. Red - loading control <u>ab8245</u> observed at 37 kDa.

ab207446 Anti-Chk2 antibody [EPR19482] was shown to specifically react with Chk2 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab264815 (knockout cell lysate ab257104) was used. Wild-type and Chk2 knockout samples were subjected to SDS-PAGE. ab207446 and Anti-alpha Tubulin antibody [EP1332Y] - Microtubule Marker (ab52866) were

incubated overnight at 4°C 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.



Western blot - Human CHEK2 (Chk2) knockout HeLa cell line (ab264815) All lanes: Anti-Chk2 antibody [EPR4325] (ab109413) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: CHEK2 knockout HeLa cell lysate

Lane 3: HEK-293 cell lysate

Lane 4: MDA-MB-231 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 61 kDa **Observed band size:** 68 kDa

Lanes 1-4: Merged signal (red and green). Green - <u>ab109413</u> observed at 68 kDa. Red - loading control <u>ab8245</u> observed at 37 kDa.

<u>ab109413</u> Anti-Chk2 antibody [EPR4325] was shown to specifically react with Chk2 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab264815 (knockout cell lysate <u>ab257104</u>) was used. Wild-type and Chk2 knockout samples were subjected to SDS-PAGE. <u>ab109413</u> and Anti-alpha Tubulin antibody [EP1332Y] - Microtubule Marker (<u>ab52866</u>) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye[®] 800CW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye[®] 680RD) preadsorbed (<u>ab216776</u>)

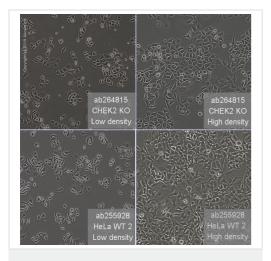
secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Mut CATITATATAAGAAAATAATTTACCTTCC<mark>C</mark>AAGAGTTTTTGACATGATGTATTCATCTCT

WT CATITATATAAGAAAATAATTTACCTTCC AAGAGTTTTTGACATGATGTATTCATCTCT

Sanger Sequencing - Human CHEK2 knockout HeLa cell line (ab264815)

Homozygous: 1 bp insertion in exon 5.



Cell Culture - Human CHEK2 (Chk2) knockout HeLa cell line (ab264815)

Representative images of CHEK2 knockout HeLa cells, low and high confluency examples (top left and right respectively) and wild-type 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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