

Anti-Chk2 antibody [EPR4325] ab109413

敲除验证 重组 RabMAb

★★★★★ [9 Abreviews](#) [21 References](#) [13 图像](#)

概述

产品名称	Anti-Chk2抗体[EPR4325]
描述	兔单克隆抗体[EPR4325] to Chk2
宿主	Rabbit
经测试应用	适用于: Flow Cyt (Intra), ICC/IF, WB, IP, IHC-P
种属反应性	与反应: Human
免疫原	Recombinant fragment within Human Chk2 aa 1-200. The exact sequence is proprietary. Database link: O96017
阳性对照	WB: HeLa (untreated and treated with gamma irradiation), HAP1, CHEK2, HEK293, MDA-MB-231, HT-29, and 293T cell lysates. IHC-P: Human colon and spleen tissues. ICC/IF: Wild-type HAP1 cells. Flow Cyt (intra): HeLa IP: HeLa whole cell lysate.
常规说明	This product is a recombinant monoclonal antibody, which offers several advantages including: <ul style="list-style-type: none">- High batch-to-batch consistency and reproducibility- Improved sensitivity and specificity- Long-term security of supply- Animal-free production For more information see here . Our RabMAb [®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents . Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.20 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant

纯度	Protein A purified
克隆	单克隆
克隆编号	EPR4325
同种型	IgG

应用

The Abpromise guarantee **Abpromise™**承诺保证使用ab109413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
Flow Cyt (Intra)		Use at an assay dependent concentration. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★★ (4)	1/100 - 1/250.
WB	★★★★★ (5)	1/50000 - 1/200000. Detects a band of approximately 62 kDa (predicted molecular weight: 61 kDa).
IP		1/10 - 1/100.
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. antigen retrieval is recommended.

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.

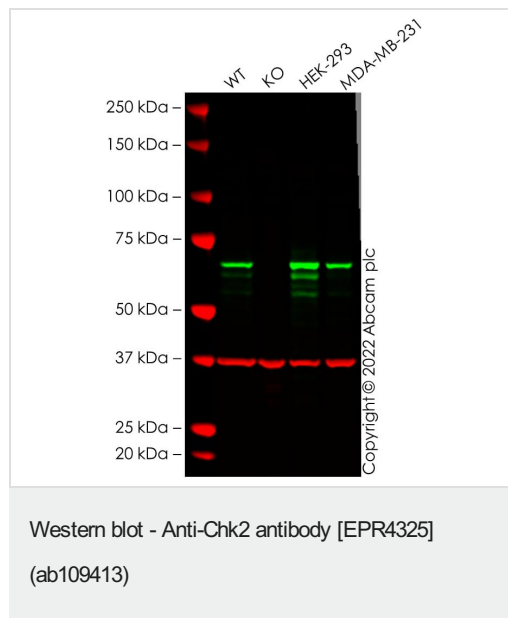
翻译后修饰

Phosphorylated by PLK4.

细胞定位

Nucleus; Nucleus. Isoform 10 is present throughout the cell and Nucleus > PML body. Nucleus > nucleoplasm. Recruited into PML bodies together with TP53.

图片



All lanes : Anti-Chk2 antibody [EPR4325] (ab109413) at 1/50000 dilution

Lane 1 : Wild-type A549 cell lysate

Lane 2 : CHEK2 knockout A549 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 IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

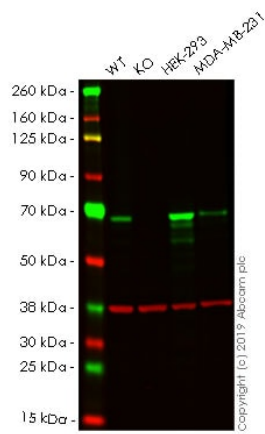
Performed under reducing conditions.

Predicted band size: 61 kDa

Observed band size: 67 kDa

False colour image of Western blot: Anti-Chk2 antibody [EPR4325] staining at 1/50000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] ([ab8245](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab109413 was shown to bind specifically to Chk2. A band was observed at 67 kDa in wild-type A549 cell lysates with no signal observed at this size in CHEK2 knockout cell line [ab276098](#) (knockout cell lysate [ab276098](#)).

To generate this image, wild-type and CHEK2 knockout A549 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-Chk2 antibody [EPR4325] (ab109413)

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.

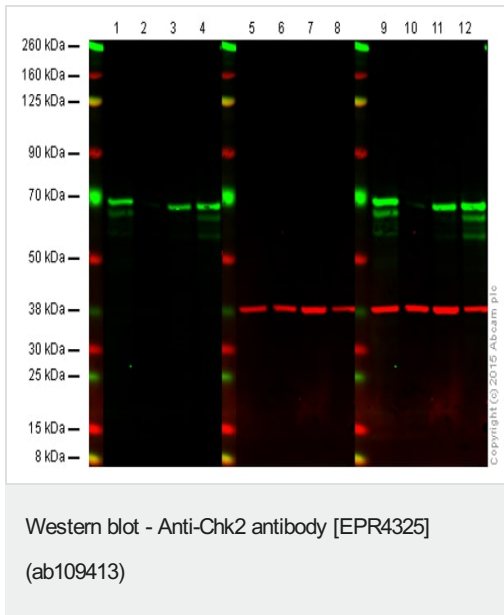
Performed under reducing conditions.

Predicted band size: 61 kDa

Observed band size: 68 kDa

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

ab109413 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 **ab257104**) was used. Wild-type and Chk2 knockout samples were subjected to SDS-PAGE. ab109413 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 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.



Lanes 1-4 : Anti-Chk2 antibody [EPR4325] (ab109413) at 1/50000 dilution

Lanes 5-8 : Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) at 1/2000 dilution

Lanes 1 & 5 : Wild-type HAP1 cell lysate

Lanes 2 & 6 : Chk2 knockout HAP1 cell lysate

Lanes 3 & 7 : HeLa cell lysate

Lanes 4 & 8 : HEK293 cell lysate

Lysates/proteins at 20 µg per lane.

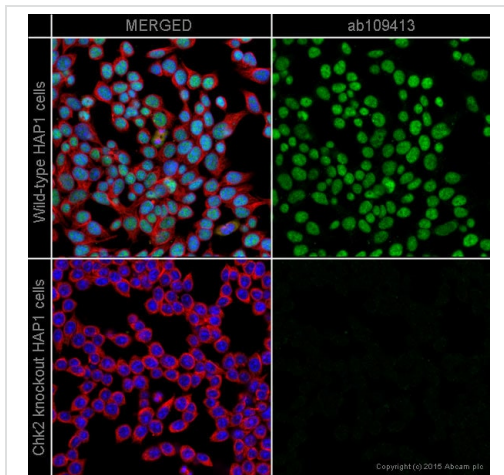
Predicted band size: 61 kDa

Lanes 1, 2, 3 and 4: Green signal from target - ab109413 observed at 62 kDa

Lanes 5, 6, 7 and 8: Red signal from loading control - **ab8245** observed at 37 kDa

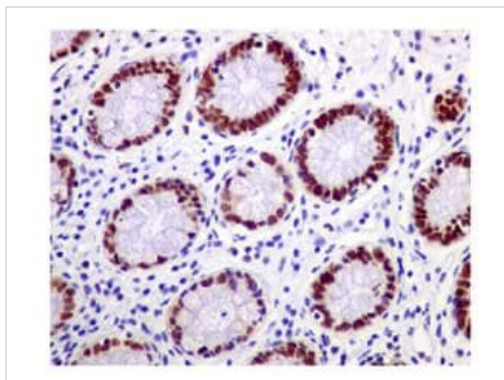
Lanes 9, 10, 11 and 12: Merged (red and green) signal

ab109413 was shown to specifically react with Chk2 when Chk2 knockout samples were used. Wild-type and Chk2 knockout samples were subjected to SDS-PAGE. ab109413 and **ab8245** (loading control to GAPDH) were diluted 1/50 000 and 1/2000 respectively and incubated overnight at 4°C. 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 1/10 000 dilution for 1 h at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-Chk2 antibody [EPR4325] (ab109413)

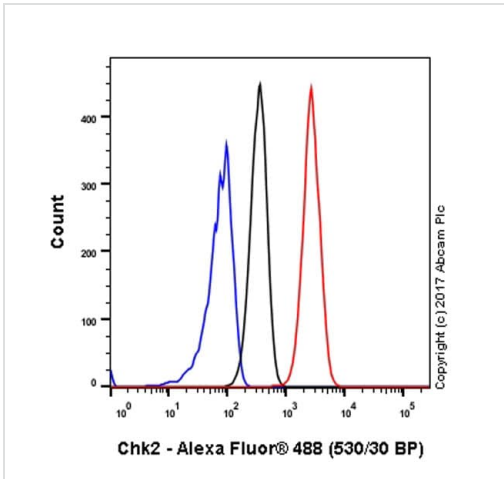
ab109413 staining Chk2 in wild-type HAP1 cells (top panel) and Chk2 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5 minutes), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1 hour. The cells were then incubated with ab109413 at 1/250 dilution and **ab195889** at 1/250 dilution (shown in pseudo colour red) overnight at +4°C, followed by a further incubation at room temperature for 1 hour with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.



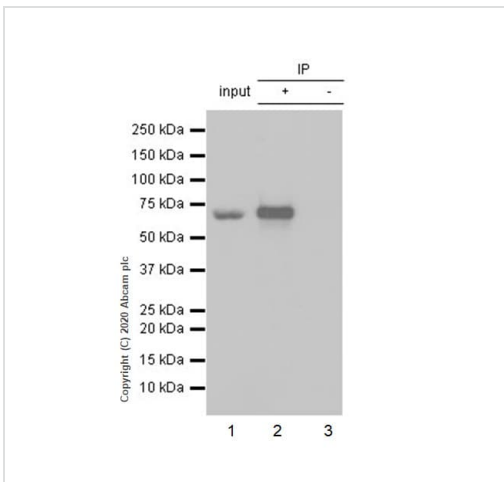
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Chk2 antibody [EPR4325] (ab109413)

Immunohistochemical analysis of paraffin-embedded human colon tissue using ab109413 at a 1/100 dilution.

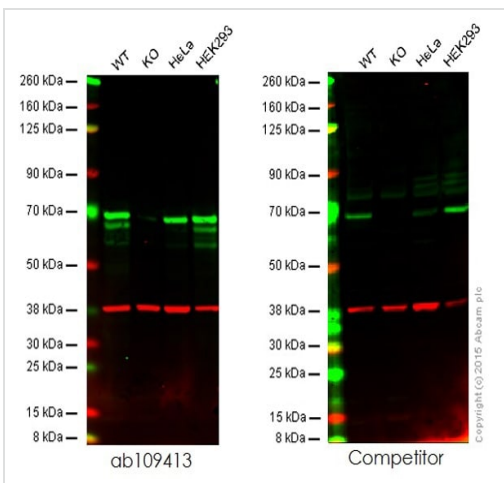
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-Chk2 antibody [EPR4325] (ab109413)



Immunoprecipitation - Anti-Chk2 antibody [EPR4325] (ab109413)



Western blot - Anti-Chk2 antibody [EPR4325] (ab109413)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling Chk2 with purified ab109413 at 1/230 dilution (10µg/ml) (red). Cells were fixed with 80% methanol and permeabilised with 0.1% Tween-20. A Goat anti rabbit IgG (Alexa Fluor® 488) (**ab150077**) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black) (**ab172730**) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.

Purified ab109413 at 1/50 dilution (2µg) immunoprecipitating Chk2 in HeLa whole cell lysate.

Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10µg

Lane 2 (+): ab109413 + HeLa whole cell lysate.

Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab109413 in HeLa whole cell lysate.

VeriBlot for IP Detection Reagent (HRP) (**ab131366**) (1/1000 dilution) was used for Western blotting.

Blocking Buffer and concentration: 5% NFDm/TBST.

Diluting buffer and concentration: 5% NFDm/TBST.

Observed band size: 62 kDa

All lanes : Anti-Chk2 antibody [EPR4325] (ab109413)

Lane 1 : Wild-type HAP1 cell lysate

Lane 2 : Chk2 knockout HAP1 cell lysate

Lane 3 : HeLa cell lysate

Lane 4 : HEK293 cell lysate

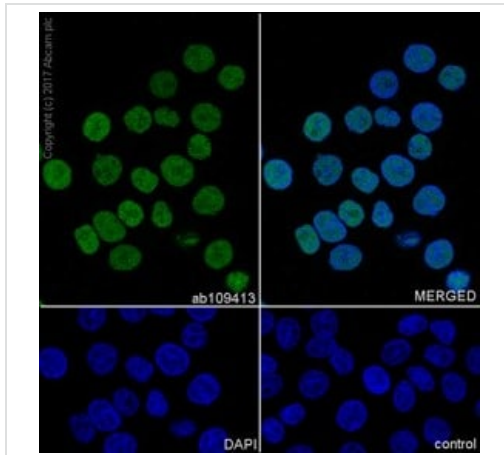
Lysates/proteins at 20 µg per lane.

Predicted band size: 61 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab109413 observed at 64 kDa. Red - loading control, **ab8245**, observed at

37 kDa.

This western blot image is a comparison between ab109413 and a competitor's rabbit polyclonal antibody.

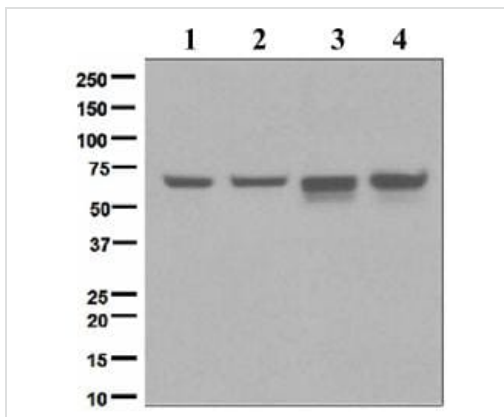


Immunocytochemistry/ Immunofluorescence - Anti-Chk2 antibody [EPR4325] (ab109413)

Immunocytochemistry analysis of HT-29 (human colorectal adenocarcinoma epithelial cell) labeling Chk2 with purified ab109413 at 1/500 dilution. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/1000 (2 µg/ml) was used as the secondary antibody. was used as counterstain.

Nuclei were stained blue with DAPI.

Negative control: PBS instead of the primary antibody.



Western blot - Anti-Chk2 antibody [EPR4325] (ab109413)

All lanes : Anti-Chk2 antibody [EPR4325] (ab109413) at 1/50000 dilution

Lane 1 : HeLa (human epithelial cell line from cervix adenocarcinoma) cells treated with gamma irradiation

Lane 2 : HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate

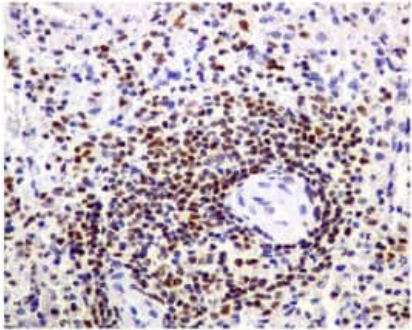
Lane 3 : HT-29 (human colorectal adenocarcinoma cell line) cell lysate

Lane 4 : HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 61 kDa

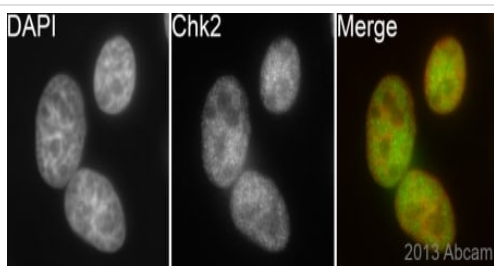
Observed band size: 62 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Chk2 antibody [EPR4325] (ab109413)

Immunohistochemical analysis of paraffin-embedded human spleen tissue using ab109413 at a 1/100 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.







Immunocytochemistry/ Immunofluorescence - Anti-Chk2 antibody [EPR4325] (ab109413)

Image courtesy of an Abreview submitted by Dr. Kirk McManus, Univ. of Manitoba/Cancer Care MCB, Canada

ab109413 (1/500) staining Chk2 in HeLa (human epithelial cell line from cervix adenocarcinoma) cells (green). Cells were fixed in paraformaldehyde, permeabilized with 0.5% Triton X-100/PBS and counterstained with DAPI in order to highlight the nucleus (red). For further experimental details please see Abreview.

Why choose a recombinant antibody?

 <p>Research with confidence Consistent and reproducible results</p>	 <p>Long-term and scalable supply Recombinant technology</p>
 <p>Success from the first experiment Confirmed specificity</p>	 <p>Ethical standards compliant Animal-free production</p>

Anti-Chk2 antibody [EPR4325] (ab109413)

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