abcam

ab108357

Anti-Cdk4 antibody [EPR4513-32-7]

Anti-Cdk4 antibody [EPR4513-32-7] (ab108357) is a rabbit monoclonal antibody detecting Cdk4 in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF**. Suitable for **Human**.

- KO validated for confirmed specificity
- Biophysical QC for unrivalled batch-batch consistency
- Over 160 publications

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR4513-32-7
Purification technique	Affinity purification Protein A
Dissociation constant	1.86 x 10^{-11} M
Concentration	0.196 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/100 - 1/250
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

WB

Tested

Species	Human
Dilution info	1/1000 - 1/10000
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/500
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/10 - 1/100
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target data

See full target information CDK4

Function Ser/Thr-kinase component of cyclin D-CDK4 (DC) complexes that phosphorylate and inhibit members of the retinoblastoma (RB) protein family including RB1 and regulate the cell-cycle during G(1)/S transition. Phosphorylation of RB1 allows dissociation of the transcription factor E2F from the RB/E2F complexes and the

subsequent transcription of E2F target genes which are responsible for the progression through the G(1) phase. Hypophosphorylates RB1 in early G(1) phase. Cyclin D-CDK4 complexes are major integrators of various mitogenenic and antimitogenic signals. Also phosphorylates SMAD3 in a cell-cycle-dependent manner and represses its transcriptional activity. Component of the ternary complex, cyclin D/CDK4/CDKN1B, required for nuclear translocation and activity of the cyclin D-CDK4 complex.

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Stable for 12 months at -20°C

Notes

What is this antibody validated in?
Anti-Cdk4 antibody [EPR4513-32-7] (ab108357) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF) in Human samples.

What is the molecular weight of Cdk4?
Anti-Cdk4 [EPR4513-32-7] (ab108357) specifically detects a band for Cdk4 (UniProt: P11802) at a molecular weight of 34kDa.

Trusted by the scientific community
Anti-Cdk4 [EPR4513-32-7] (ab108357) was first used in a scientific publication in 2011 and has been cited over 160 times in peer-reviewed journals.

Specificity confirmed
The specificity of Anti-Cdk4 antibody [EPR4513-32-7] (ab108357) has been confirmed by Western blot testing in CDK4 Knockout HAP1 cells.

Other related products
We have a range of other formats of antibody clone [EPR4513-32-7] also available for your convenience: ab108357, Alexa Fluor® 488 - ab193966, Alexa Fluor® 647 - ab193967, HRP - ab193968, Carrier free - ab213216, Alexa Fluor® 555 - ab215318

Species reactivity
Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Cdk4 also known as Cyclin-dependent kinase 4 is a serine/threonine kinase involved in cell cycle regulation. Cdk4 has a molecular weight of approximately 34 kDa. This protein mainly locates in the nucleus of the cell and its expression occurs in diverse tissues. Cdk4 partners with D-type cyclins to exert its function which is essential for the transition from the G1 phase to the S phase of the cell cycle. Researchers often study Cdk4 using techniques like immunohistochemistry (IHC) to determine its expression and localization in different tissues.
Biological function summary	Cdk4 plays a fundamental role in cell cycle regulation by forming a complex with D-type cyclins. Together they phosphorylate the retinoblastoma protein (Rb) resulting in the release of E2F transcription factors which promote the expression of genes necessary for DNA replication. The Cdk4/cyclin D complex regulates the cell's commitment to enter S phase and continue cell division. This regulation is key for normal cell proliferation and tissue homeostasis. Cdk4 activity is strictly controlled by INK4 family members acting as inhibitors and adding an extra regulation layer.
Pathways	Cdk4 is an integral part of important signaling pathways like the cell cycle and PI3K/AKT pathways. In the cell cycle pathway Cdk4 relays signals downstream that drive the transition from the G1 to S phase through its interaction with cyclin D and Rb. In the PI3K/AKT pathway signaling can influence cyclin D expression indirectly affecting Cdk4 activity. Proteins such as Cdk6 closely relate to Cdk4 and often compensate or partner with Cdk4 in these pathways to maintain proper cell cycle progression.
Associated diseases and disorders	Abnormalities in Cdk4 function or regulation relate to cancer and certain metabolic syndromes. Hyperactivity of Cdk4 due to overexpression or mutation

frequently occurs in several cancers including melanoma and breast cancer. In these malignancies Cdk4 aberrantly drives excessive cell proliferation. Mutations or alterations in proteins such as p16 an inhibitor of Cdk4 are often found in these cancers highlighting the critical role of Cdk4 in disease progression. Understanding the role of Cdk4 allows researchers to develop targeted therapies such as Cdk4/6 inhibitors offering new treatment options.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

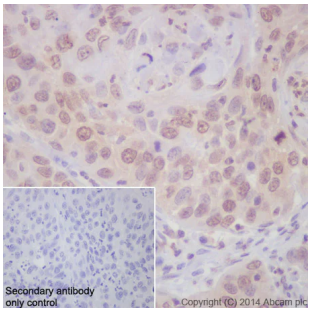
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

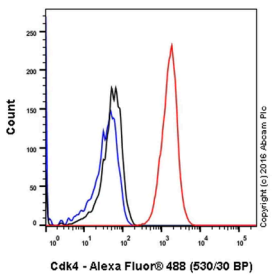
Full details and terms and conditions can be found here:
Terms & Conditions.

11 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervix carcinoma tissue labeling Cdk4 with purified ab108357 at 1/100. Heat mediated antigen retrieval was performed using Tris/EDTA buffer (pH 9). ab97051, a HRP-conjugated goat anti-rabbit IgG (H+L), was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

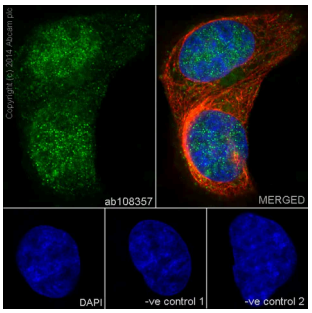


Flow Cytometry (Intracellular) - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Cdk4 Flow Cytometry (Intracellular) staining using rabbit Anti-Cdk4 antibody

ab108357 staining CDK4in the human cell line MCF7 (human breast adenocarcinoma cell line) by intracellular flow cytometry. Cells were fixed with 4% paraformaldehyde, permeabilized with 90% methanol and the sample was incubated with the primary antibody at a dilution of 1/20. A goat anti rabbit IgG (Alexa Fluor® 488) at a dilution of 1/2000 was used as the secondary antibody. Isoytype control: Rabbit monoclonal IgG (Black)

Unlabeled control: Cell without incubation with primary antibody and secondary antibody (Blue)

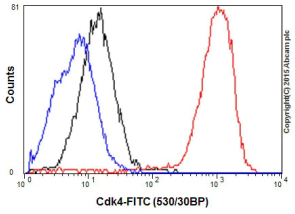


Immunocytochemistry/ Immunofluorescence - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Cdk4 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Cdk4 antibody

Immunocytochemical analysis of MCF7 (human breast adenocarcinoma cell line) cells, labeling Cdk4 with purified ab108357 at 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500), was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. ab7291, a mouse anti-tubulin (1/1000) and ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500) were also used.
Control 1: primary antibody (1/100) and secondary antibody, ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500).

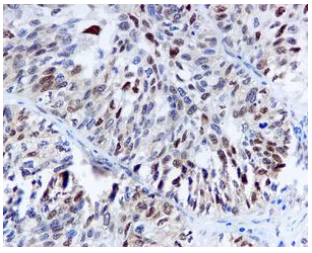
Control 2: ab7291 (1/1000) and secondary antibody, ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).



Flow Cytometry (Intracellular) - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Cdk4 Flow Cytometry (Intracellular) staining using rabbit Anti-Cdk4 antibody

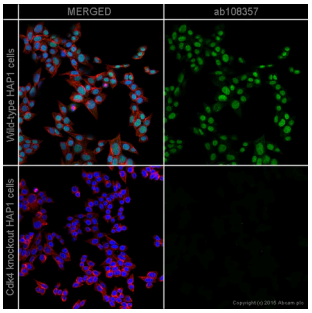
Intracellular Flow Cytometry analysis of MCF7 (human breast adenocarcinoma cell line) cells labelling Cdk4 with purified ab108357 at 1/40 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Cdk4 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Cdk4 antibody

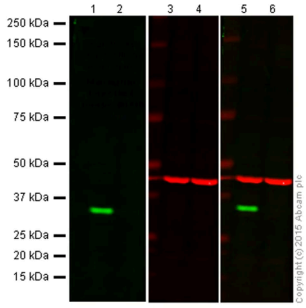
Immunohistochemical analysis of formalin/PFA-fixed paraffin-embedded human urothelial carcinoma tissue labelling Cdk4 with unpurified ab108357 at a dilution of 1/100.
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Cdk4 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Cdk4 antibody

ab108357 staining Cdk4 in wild-type HAP1 cells (top panel) and Cdk4 knockout HAP1 cells (bottom panel). The cells were fixed with 4% formaldehyde (10min), 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 1h. The cells were then incubated with ab108357 at 1/500 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 1h 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.



Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Cdk4 Western blot staining using rabbit Anti-Cdk4 antibody

Lanes 5 and 6: Merged signal (red and green). Green - ab108357 observed at 34kDa. Red - loading control to beta actin observed at 40kDa.
ab108357 was shown to specifically react with CDK4 in wild-type HAP1 cells. No band was observed when CDK4 knockout samples were examined. Wild-type and CDK4 knockout samples were subjected to SDS-PAGE. ab108357 and ab8226 (loading control to beta actin) were both diluted at 1/1000 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 hour at room temperature before imaging.

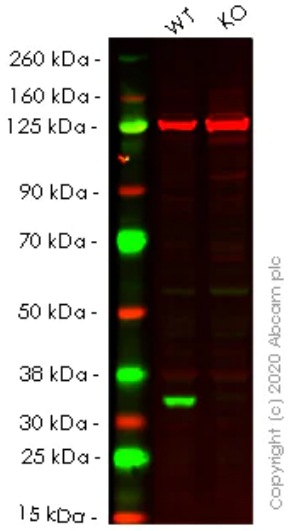
Lanes 1 - 4:
Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357) at 1/1000 dilution

Lanes 5 - 6:
Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Lanes 1, 3 and 5:
Wild-type HAP1 cell lysate at 20 µg

Lanes 2, 4 and 6:
CDK4 knockout HAP1 cell lysate at 20 µg

Predicted band size: 34 kDa



Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Cdk4 Western blot staining using rabbit Anti-Cdk4 antibody

Lanes 1- 2: Merged signal (red and green). Green - ab108357 observed at 34 kDa. Red - Anti-Vinculin antibody [VIN-54] observed at 124 kDa.
ab108357 was shown to react with Cdk4 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab255378 (knockout cell lysate ab263780) was used. Wild-type HeLa and CDK4 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. ab108357 and Anti-Vinculin antibody [VIN-54] overnight at 4°C at a 1 in 1000 dilution and a 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.

All lanes:
Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357) at 1/1000 dilution

Lane 1:
Wild-type HeLa cell lysate at 20 µg

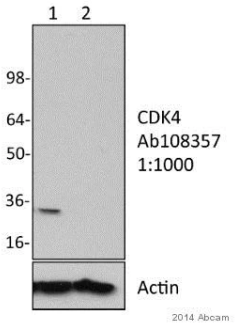
Lane 2:
CDK4 knockout HeLa cell lysate at 20 µg

Lane 2:
Western blot - Human CDK4 knockout HeLa cell line (ab255378)

Performed under reducing conditions.

Predicted band size: 34 kDa, 55 kDa

Observed band size: 34 kDa, 55 kDa



This image is courtesy of a customer review submitted by Sonia Rocha

Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

All lanes:
Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357) at 1/1000 dilution

Lane 1:
Human osteosarcoma whole cell lysate - control, non-targeting siRNA at 20 µg

Lane 2:
Human osteosarcoma whole cell lysate - siRNA for CDK4 at 20 µg

Secondary

All lanes:
HRP-conjugated goat anti-rabbit IgG polyclonal at 1/2000 dilution

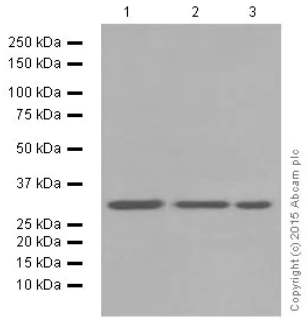
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 34 kDa

Observed band size: 34 kDa

Exposure time: 5s



Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

Blocking and dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357) at 1/10000 dilution

Lane 1:
HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate at 20 µg

Lane 2:
K562 (human chronic myelogenous leukemia cell line from bone marrow) cell lysate at 20 µg

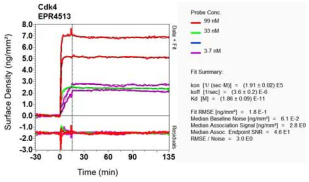
Lane 3:
Ramos (human Burkitt's lymphoma cell line) cell lysate at 20 µg

Secondary

All lanes:
Peroxidase-conjugated goat anti-rabbit IgG, (H+L) at 1/1000 dilution

Predicted band size: 34 kDa

Observed band size: 34 kDa



OI-RD Scanning - Anti-Cdk4 antibody [EPR4513-32-7] (ab108357)

We have systematically measured KD (the equilibrium dissociation constant between the antibody and its antigen), of more than 840 recombinant antibodies to assess not only their individual KD values but also to see the average affinity of antibody. Based on the comparison with published literature values for mouse monoclonal antibodies, Recombinant antibodies appear to be on average 1-2 order of magnitude higher affinity.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.