abcam

ab32444

Anti-Cdc25C antibody [E302]

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	E302
Purification technique	Affinity purification Protein A
Specificity	The antibody can also detect splice isoform 2, 4 and 5 of human Cdc25C, based on sequence homology.
Concentration	0.995 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	1/30 - 1/80

Notes	-

WB

Tested

Species	Human
Dilution info	1/1000 - 1/5000
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/250 - 1/500
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/2500
Notes	**For unpurified, use 1/250 - 1/500.**

Target data

See full target information CDC25C

Function	Functions as a dosage-dependent inducer in mitotic control. Tyrosine protein phosphatase required for progression of the cell cycle (PubMed:8119945). When phosphorylated, highly effective in activating G2 cells into prophase (PubMed:8119945). Directly dephosphorylates CDK1 and activates its kinase activity (PubMed:8119945).

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	Avoid freeze / thaw cycle

Notes

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.

Supplementary info

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

Activity summary	Cdc25C also known as dual specificity phosphatase Cdc25C is a 65-kDa protein that acts as a cell cycle regulator controlling the transition from the G2 to M phase. The protein is expressed in various tissues but has higher expression levels in actively dividing cells. Cdc25C is important for cell division due to its ability to activate cyclin-dependent kinases (CDKs) by dephosphorylating the inactive phosphorylated forms. This action makes Cdc25C an important target for regulating the cell cycle and ensuring proper cell proliferation.
Biological function summary	The proper function of Cdc25C involves facilitating proper cell cycle progression by coordinating with other cell cycle regulators. It participates in a complex network where it dephosphorylates and activates CDK1/cyclin B1 complexes promoting the mitotic entry. This function is important for maintaining genomic stability during cell division. Misregulation of Cdc25C can lead to cell cycle arrest or uncontrolled cell proliferation highlighting its essential role in cell cycle control mechanisms.
Pathways	Cdc25C fits into the cell cycle checkpoint pathways and is also a part of the DNA damage response pathways. It connects with the p53 and ATM/ATR signaling proteins while responding to DNA damage ensuring a temporary pause in cell cycle progression for repair mechanisms to act. Proper interaction with these pathways is essential for maintaining cellular integrity and preventing the proliferation of damaged cells with CDK1 and Wee1 kinase serving as major interacting proteins in these processes.
Associated diseases and disorders	Misregulation or overexpression of Cdc25C can relate to cancer and various cell proliferation disorders. In cancer Cdc25C's disordered expression often links to unchecked cell division and tumor progression commonly involving proteins such as p53 which act as tumor suppressors. Furthermore abnormalities in Cdc25C function may lead to issues related to cell cycle progression errors implicating it in other proliferative disorders such as hyperplasia where it acts in conjunction with and alters the activities of proteins like cyclin B1.

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

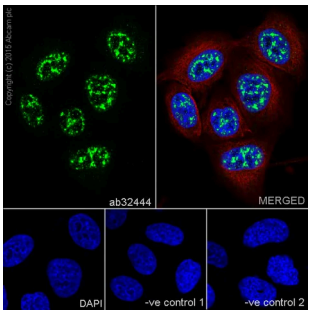
We do not recommend this combination. It is not covered by our product promise.

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.

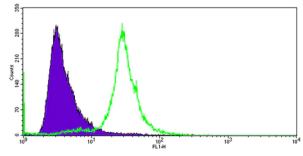
11 product images



Immunocytochemistry/ Immunofluorescence - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Cdc25C antibody

Immunocytochemistry/Immunofluorescence analysis of HeLa (human cervix adenocarcinoma) cells labelling Cdc25C with purified ab32444 at 1/400. Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. The cells were co-stained with ab7291, a mouse anti-tubulin antibody (1/1000) using ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000) as the secondary. Nuclei couterstained with DAPI (blue).
For negative control 1, rabbit primary antibody was used, followed by anti-mouse secondary antibody (ab150120). For negative control 2, mouse primary antibody (ab7291) was used followed by anti-rabbit secondary antibody (ab150077).

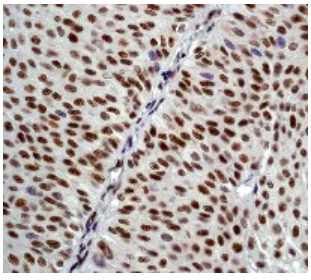


This image is courtesy of a customer review submitted by Brandon White.

Flow Cytometry (Intracellular) - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Flow Cytometry (Intracellular) staining using rabbit Anti-Cdc25C antibody

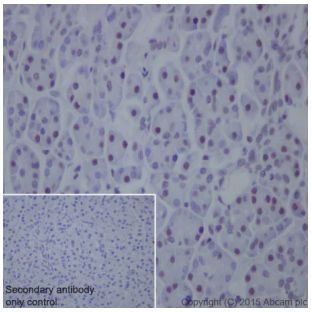
Intracellular Flow Cytometry analysis of HeLa cells, staining Cdc25C with unpurified ab32444. Cells were fixed with formaldehyde and permeabilized with 90% methanol. Samples were incubated with primary antibody (1/20 in PBS + 10% goat serum) for 1 hour at 23°C. A FITC-conjugated goat anti-rabbit polyclonal IgG (1/1000) was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Cdc25C antibody

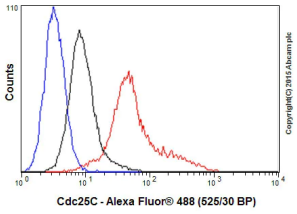
Immunohistochemical analysis of paraffin-embedded human urinary bladder carcinoma unpurified ab32444 at 1/250 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Cdc25C antibody

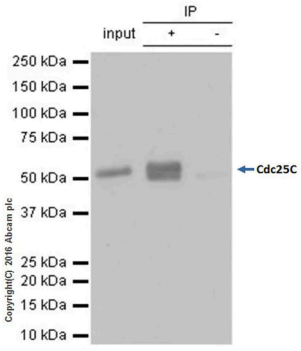
Immunohistochemical analysis of paraffin embedded human pancreas tissue section labelling Cdc25C with purified ab32444 at dilution of 1/2500. The secondary antibody used was Goat Anti-Rabbit IgG H&L (HRP) (ab97051), at dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.



Flow Cytometry (Intracellular) - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Flow Cytometry (Intracellular) staining using rabbit Anti-Cdc25C antibody

Intracellular Flow Cytometry analysis of K562 (human chronic myelogenous leukemia) cells labelling Cdc25C with purified ab32444 at 1/180 (red). Cells were fixed with 4% paraformaldehyde. Alexa Fluorr® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Immunoprecipitation - Anti-Cdc25C antibody [E302] (ab32444)

ab32444 (purified) at 1/30 immunoprecipitating Cdc25C in HeLa (human cervix adenocarcinoma) whole cell lysate.
Lane 1 (input): HeLa (human cervix adenocarcinoma) whole cell lysate

Lane 2 (+): ab32444 + HeLa (human cervix adenocarcinoma) whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab32444 in HeLa (human cervix adenocarcinoma) whole cell lysate

For western blotting, ab131366 VeriBlot for IP Detection Reagent (HRP) was used for detection (1/10000).

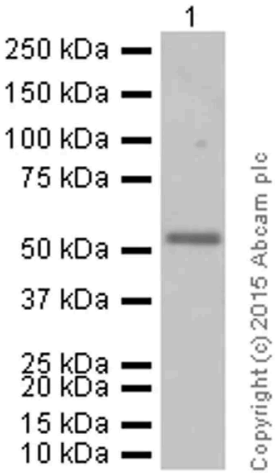
Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

All lanes:
Immunoprecipitation - Anti-Cdc25C antibody [E302] (ab32444)

Predicted band size: 53 kDa

Observed band size: 60 kDa



Western blot - Anti-Cdc25C antibody [E302] (ab32444)

Blocking and diluting buffer 5% NFDM/TBST
All lanes:
Western blot - Anti-Cdc25C antibody [E302] (ab32444) at 1/1000 dilution

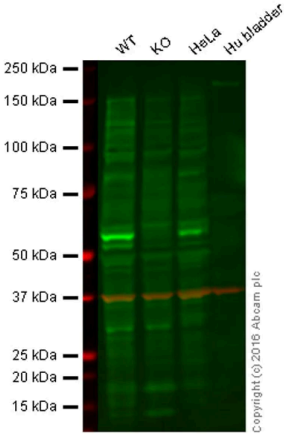
All lanes:
HeLa (human cervix adenocarcinoma) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 53 kDa

Observed band size: 60 kDa



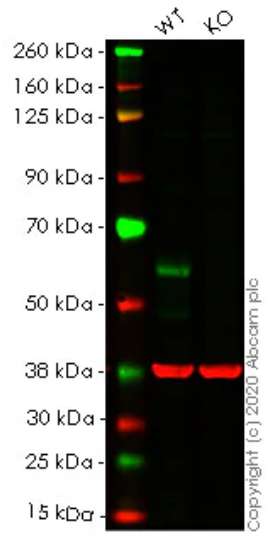
Western blot - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Western blot staining using rabbit Anti-Cdc25C antibody

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: Cdc25C knockout HAP1 cell lysate (20 µg)
Lane 3: HeLa cell lysate (20 µg)
Lane 4: Hu bladder cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab32444 observed at 55 kDa. Red - loading control, ab8245, observed at 37 kDa.
ab32444 was shown to recognize Cdc25C when Cdc25C knockout samples were used, along with additional cross-reactive bands. Wild-type and Cdc25C knockout samples were subjected to SDS-PAGE. ab32444 and ab8245 (loading control to GAPDH) were diluted at 1/2500 and 1/10 000 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 1/10000 dilution for 1 h at room temperature before imaging.

All lanes:
Western blot - Anti-Cdc25C antibody [E302] (ab32444)

Predicted band size: 53 kDa



Western blot - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Western blot staining using rabbit Anti-Cdc25C antibody

Lanes 1- 2: Merged signal (red and green). Green - ab32444 observed at 58 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.
ab32444 was shown to react with Cdc25C in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265189 (knockout cell lysate ab257387) was used. Wild-type HeLa and CDC25C 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. ab32444 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated 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-Cdc25C antibody [E302] (ab32444) at 1/1000 dilution

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

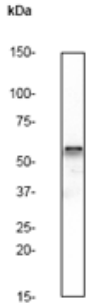
Lane 2:
CDC25C knockout HeLa cell lysate at 20 µg

Lane 2:
Western blot - Human CDC25C knockout HeLa cell line (ab265189)

Performed under reducing conditions.

Predicted band size: 53 kDa

Observed band size: 58 kDa



Western blot - Anti-Cdc25C antibody [E302] (ab32444)

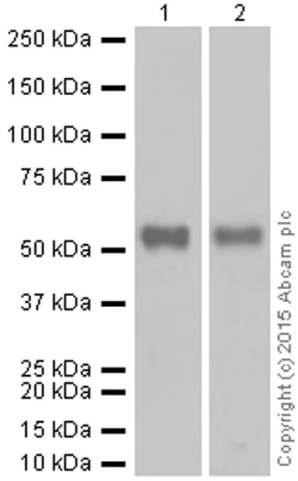
Cdc25C Western blot staining of HeLa cell lysate using rabbit Anti-Cdc25C antibody

All lanes:
Western blot - Anti-Cdc25C antibody [E302] (ab32444) at 1/5000 dilution

All lanes:
HeLa cell lysate

Predicted band size: 53 kDa

Observed band size: 60 kDa



Western blot - Anti-Cdc25C antibody [E302] (ab32444)

Cdc25C Western blot staining using rabbit Anti-Cdc25C antibody

Blocking and diluting buffer 5% NFDM/TBST
All lanes:
Western blot - Anti-Cdc25C antibody [E302] (ab32444) at 1/5000 dilution

Lane 1:
K562 (human chronic myelogenous leukemia) whole cell lysate at 20 µg

Lane 2:
HEK293 (human embryonic kidney) whole cell lysates at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 53 kDa

Observed band size: 60 kDa

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