abcam

Product datasheet

Anti-CDC42 antibody [EPR15620] ab187643

KO VALIDATED Recombinant RabMAb

1 Abreviews 43 References 12 Images

Overview

Product name	Anti-CDC42 antibody [EPR15620]
Description	Rabbit monoclonal [EPR15620] to CDC42
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, IP, WB, Flow Cyt (Intra), ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HEK-293T, HepG2, MCF7, HT-29, Jurkat and HeLa whole cell lysate (**ab150035**); Mouse brain, Mouse kidney, Mouse spleen and Rat spleen lysates. IP: HT-29 cell lysate. Flow Cyt (intra): HeLa cells. ICC/IF: U937 cells. IHC-P: Human lung carcinoma, breast carcinoma and colon tissues; Rat colon tissue.
General notes	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 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**. **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.**

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2

	Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR15620
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab187643 in the following tested applications.

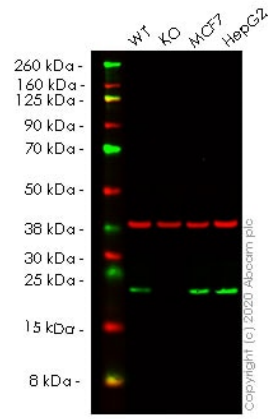
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P		1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		1/20 - 1/40.
WB		1/10000 - 1/50000. Detects a band of approximately 21 kDa (predicted molecular weight: 21 kDa).
Flow Cyt (Intra)		1/170. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★☆ (1)	1/500.

Target

Function	Plasma membrane-associated small GTPase which cycles between an active GTP-bound and an inactive GDP-bound state. In active state binds to a variety of effector proteins to regulate cellular responses. Involved in epithelial cell polarization processes. Causes the formation of thin, actin-rich surface projections called filopodia.
Sequence similarities	Belongs to the small GTPase superfamily. Rho family. CDC42 subfamily.
Post-translational modifications	AMPylation at Tyr-32 and Thr-35 are mediated by bacterial enzymes in case of infection by H.somnus and V.parahaemolyticus, respectively. AMPylation occurs in the effector region and leads to inactivation of the GTPase activity by preventing the interaction with downstream effectors, thereby inhibiting actin assembly in infected cells. It is unclear whether some human enzyme mediates AMPylation; FICD has such ability in vitro but additional experiments remain to be done to confirm results in vivo.
Cellular localization	Cell membrane.
Form	There are 2 isoforms produced by alternative splicing. Isoform 1 also known as: Brain; Isoform 2 also known as: Placental.

Images



Western blot - Anti-CDC42 antibody [EPR15620] (ab187643)

All lanes : Anti-CDC42 antibody [EPR15620] (ab187643) at 1/5000 dilution

Lane 1 : Wild-type HEK-293T cell lysate
Lane 2 : CDC42 knockout HEK-293T cell lysate
Lane 3 : MCF7 cell lysate
Lane 4 : HepG2 cell lysate

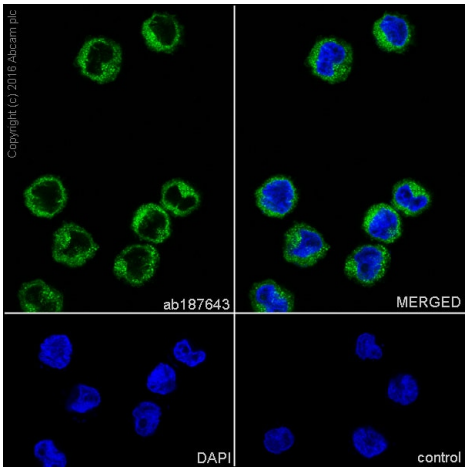
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 21 kDa

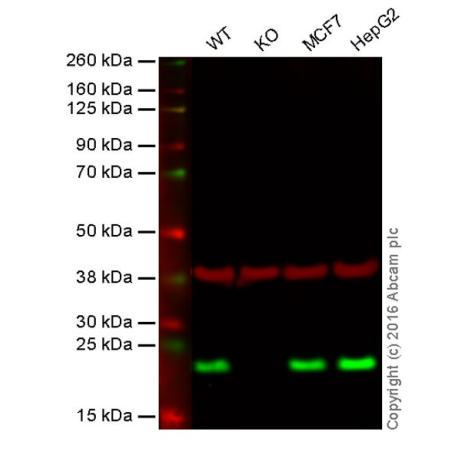
Lanes 1-4: Merged signal (red and green). Green - ab187643 observed at 20 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab187643 Anti-CDC42 antibody [EPR15620] was shown to specifically react with CDC42 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line **ab266522** (knockout cell lysate **ab256868**) was used. Wild-type and CDC42 knockout samples were subjected to SDS-PAGE. ab187643 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 5000 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.



Immunocytochemistry/ Immunofluorescence - Anti-CDC42 antibody [EPR15620] (ab187643)

Immunocytochemistry/Immunofluorescence analysis of U937 (Human histiocytic lymphoma cell line) labelling CDC42 with purified ab187643 at 1/500. Cells were fixed with 4% PFA and permeabilized with 0.1% triton X-100. **ab150077** Goat anti rabbit IgG (Alexa Fluor® 488) at 1/1000 was used as the secondary antibody. Nuclei were counterstained with DAPI. PBS was used instead of the primary antibody as the negative control.



Western blot - Anti-CDC42 antibody [EPR15620] (ab187643)

All lanes : Anti-CDC42 antibody [EPR15620] (ab187643) at 1/5000 dilution

Lane 1 : Wild-type HAP1 cell lysate
Lane 2 : CDC42 knockout HAP1 cell lysate
Lane 3 : MCF7 cell lysate
Lane 4 : HepG2 cell lysate

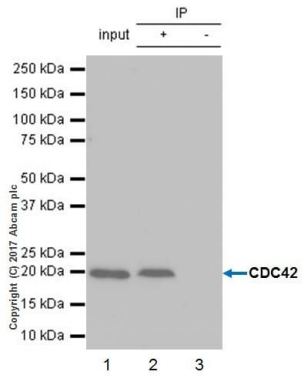
Lysates/proteins at 20 µg per lane.

Predicted band size: 21 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab187643 observed at 20 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab187643 was shown to specifically react with CDC42 in wild-type HAP1 cells. No band was observed when CDC42 knockout

samples were examined. Wild-type and CDC42 knockout samples were subjected to SDS-PAGE. ab187643 and **ab8245** (loading control to GAPDH) were diluted at 1/5000 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 1/10,000 dilution for 1 hour at room temperature before imaging.

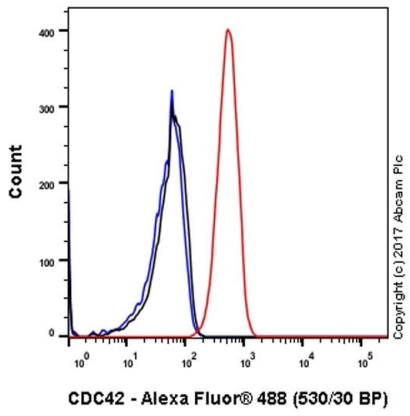


Immunoprecipitation - Anti-CDC42 antibody [EPR15620] (ab187643)

Lane 1 (input): HT-29 (human colorectal adenocarcinoma epithelial cell) whole cell lysate,10µg
Lane 2(+): HT-29 whole cell lysate
Lane 3(-): Rabbit monoclonal IgG (**ab172730**) instead of ab187643 in HT-29 whole cell lysate

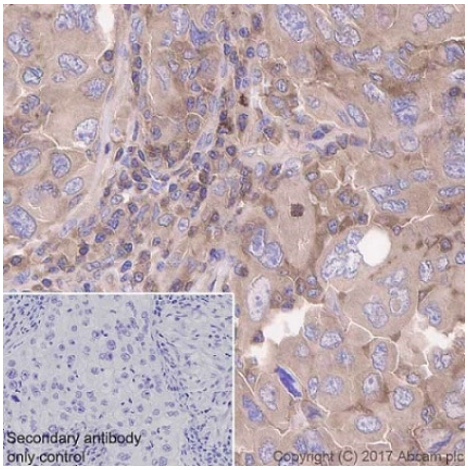
Ab187643 immunoprecipitating CDC42 in HT-29 whole cell lysate. Capture antibody was used at a 1:60 dilution. For western blotting, ab187643 was used as the primary antibody at a 1:1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST



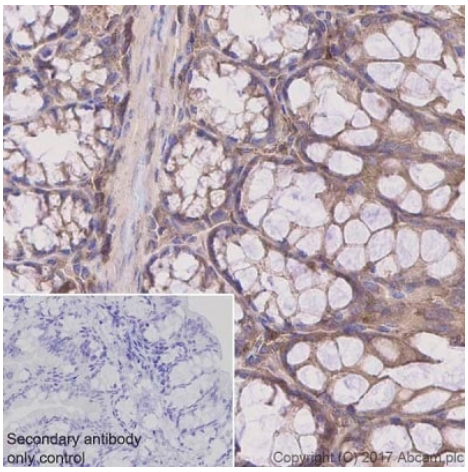
Flow Cytometry (Intracellular) - Anti-CDC42 antibody [EPR15620] (ab187643)

Intracellular Flow Cytometry analysis of HeLa cells (human cervix adenocarcinoma epithelial).Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Primary antibody was used at a 1/120 dilution (red). A Goat anti rabbit IgG (Alexa Fluor®488, **ab150077**) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (**ab172730**) was used as the isotype control (black). Cell without incubation with primary antibody and secondary antibody (Blue).



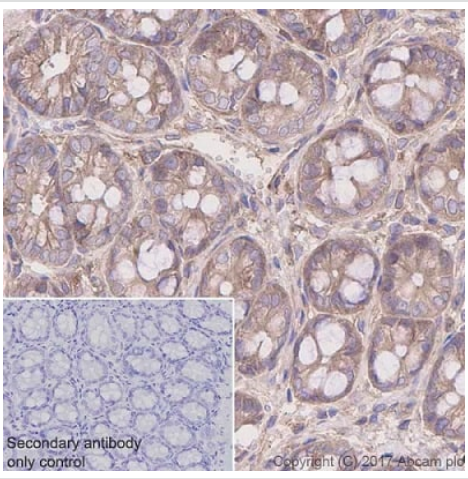
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CDC42 antibody [EPR15620] (ab187643)

ab187643 staining CDC42 in Human lung carcinoma tissue sections by Immunohistochemistry (IHC-P- paraformaldehyde-fixed, paraffin-embedded sections). Antigen retrieval was by heat mediation using **ab93684** (Tris/EDTA buffer, pH 9.0). Samples were incubated with primary antibody at a 1/500 dilution. A ready to use Goat Anti-Rabbit IgG H&L (HRP) was used as the secondary antibody. Hematoxylin was used as a counter stain. Cytoplasmic staining on human lung carcinoma.



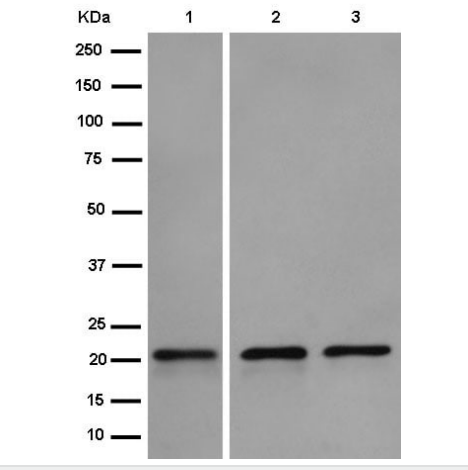
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CDC42 antibody [EPR15620] (ab187643)

ab187643 staining CDC42 in Mouse colon tissue sections by Immunohistochemistry (IHC-P- paraformaldehyde-fixed, paraffin-embedded sections). Antigen retrieval was by heat mediation using **ab93684** (Tris/EDTA buffer, pH 9.0). Samples were incubated with primary antibody at a 1/500 dilution. A ready to use Goat Anti-Rabbit IgG H&L (HRP) was used as the secondary antibody. Hematoxylin was used as a counter stain. Cytoplasmic staining on mouse colon.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CDC42 antibody [EPR15620] (ab187643)

ab187643 staining CDC42 in Rat colon tissue sections by Immunohistochemistry (IHC-P- paraformaldehyde-fixed, paraffin-embedded sections). Antigen retrieval was by heat mediation using **ab93684** (Tris/EDTA buffer, pH 9.0). Samples were incubated with primary antibody at a 1/500 dilution. A ready to use Goat Anti-Rabbit IgG H&L (HRP) was used as the secondary antibody. Hematoxylin was used as a counter stain. Cytoplasmic staining on rat colon.



Western blot - Anti-CDC42 antibody [EPR15620] (ab187643)

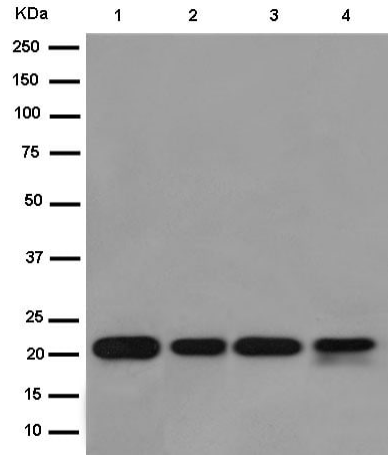
All lanes : Anti-CDC42 antibody [EPR15620] (ab187643) at 1/20000 dilution

Lane 1 : Jurkat cell lysate
Lane 2 : HT-29 cell lysate
Lane 3 : HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugate at 1/1000 dilution

Predicted band size: 21 kDa
Observed band size: 21 kDa



Western blot - Anti-CDC42 antibody [EPR15620] (ab187643)

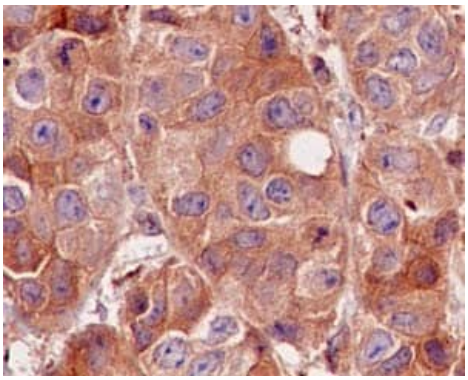
All lanes : Anti-CDC42 antibody [EPR15620] (ab187643) at 1/5000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse kidney lysate
Lane 3 : Mouse spleen lysate
Lane 4 : Rat spleen lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugate at 1/1000 dilution

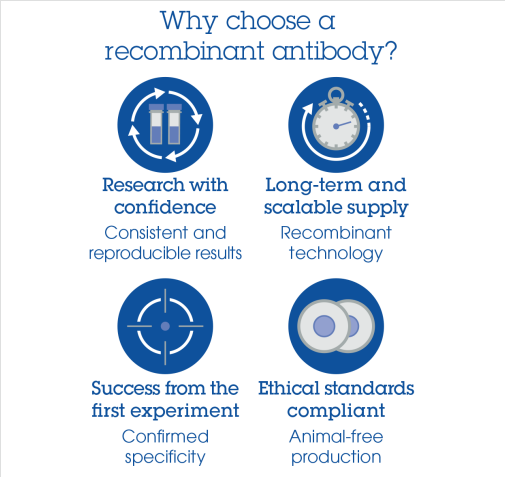
Predicted band size: 21 kDa
Observed band size: 21 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CDC42 antibody [EPR15620] (ab187643)

Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue labeling CDC42 with ab187643 at 1/250 dilution, followed by prediluted HRP Polymer for Rabbit IgG. Counter stained with Hematoxylin.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Anti-CDC42 antibody [EPR15620] (ab187643)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.com/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors