abcam

ab222487

Anti-DUSP4 antibody [EPR19881] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR19881
Purification technique	Affinity purification Protein A
Concentration	1.053 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	-

Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	This product gave a positive signal in A549 (DUSP4 knockout A549 cells used as a negative control) fixed with 100% methanol (5 min).

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-

Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information DUSP4

Function	Regulates mitogenic signal transduction by dephosphorylating both Thr and Tyr residues on MAP kinases ERK1 and ERK2.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab222487 is the carrier-free version of ab216576.

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.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

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

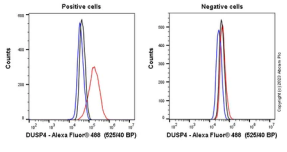
We do not recommend this combination. It is not covered by our product promise.

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.

8 product images



Flow Cytometry (Intracellular) - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

DUSP4 Flow Cytometry (Intracellular) staining using rabbit Anti-DUSP4 antibody

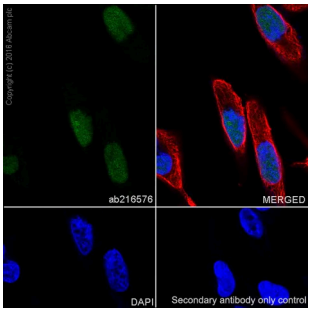
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab216576).

Flow cytometry overlay histogram showing left wild-type A549 positive cells and right negative DUSP4 knockout A549 stained with ab216576 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab216576) (1x 106 in 100µl at 1.0 µg/ml (1/1990)) for 30min at 22°C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C

Isotype control antibody Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (black line) was used at the same concentration and conditions as the primary antibody. Unlabelled sample was also used as a control (blue line).

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.



Immunocytochemistry/ Immunofluorescence - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

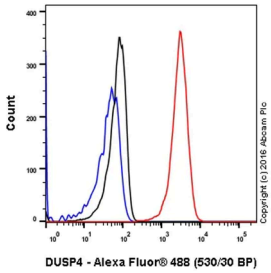
DUSP4 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-DUSP4 antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized MDA-MB-231 (Human breast adenocarcinoma cell line) cells labeling DUSP4 with ab216576 at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).
Confocal image showing nuclear staining on MDA-MB-231 cell line.

The nuclear counterstain is DAPI (blue). Tubulin is detected with ab195889 (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/1000 dilution.

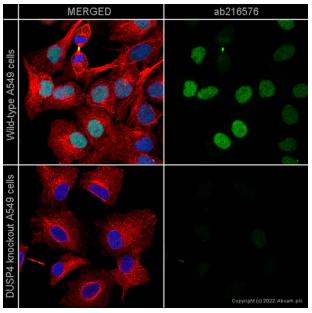
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab216576).



Flow Cytometry (Intracellular) - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

DUSP4 Flow Cytometry (Intracellular) staining using rabbit Anti-DUSP4 antibody

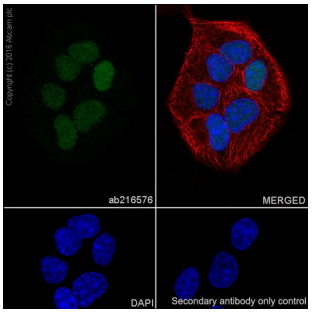
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed MDA-MB-231 (Human breast adenocarcinoma cell line) cells labeling DUSP4 with ab216576 at 1/60 dilution (red) compared with a rabbit monoclonal IgG isotype control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab216576).



Immunocytochemistry/ Immunofluorescence - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

DUSP4 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-DUSP4 antibody

ab216576 staining DUSP4 in wild-type A549 cells, with negative expression in DUSP4 knockout A549 cells. The cells were fixed with 100% methanol (5 min), permeabilised 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 overnight at +4°C with ab216576 at 1 µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 µg/ml. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150119, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab216576).



Immunocytochemistry/ Immunofluorescence - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

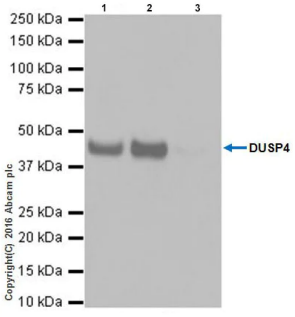
DUSP4 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-DUSP4 antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A549 (Human lung carcinoma cell line) cells labeling DUSP4 with ab216576 at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).
Confocal image showing nuclear staining on A549 cell line.

The nuclear counterstain is DAPI (blue). Tubulin is detected with ab195889 (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/1000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab216576).



Immunoprecipitation - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

DUSP4 was immunoprecipitated from 0.35mg of MDA-MB-231 (Human breast adenocarcinoma cell line) whole cell lysate with ab216576 at 1/30 dilution.
Western blot was performed from the immunoprecipitate using ab216576 at 1/500 dilution.

VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1000 dilution.

Lane 1: MDA-MB-231 whole cell lysate, 10µg (Input).

Lane 2: ab216576 IP in MDA-MB-231 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab216576 in MDA-MB-231 whole cell lysate.

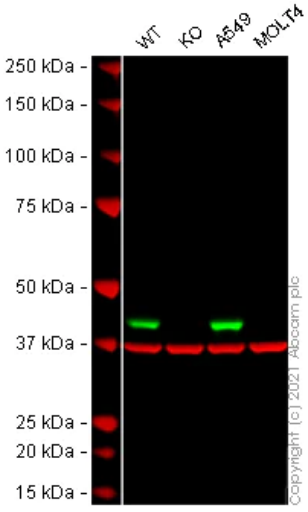
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab216576).
All lanes:
Immunoprecipitation - Anti-DUSP4 antibody [EPR19881] (ab216576)

Predicted band size: 43 kDa

Observed band size: 43 kDa



Western blot - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

DUSP4 Western blot staining using rabbit Anti-DUSP4 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab216576).
Lanes 1 - 4: Merged signal (red and green). Green - ab216576 observed at 40 kDa. Red - loading control ab8245 (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab216576 was shown to react with DUSP4 in wild-type A549 cells in Western blot with loss of signal observed in DUSP4 knockout cell line ab273859 (DUSP4 knockout cell lysate ab273813). Wild-type A549 and DUSP4 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab216576 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.

All lanes:
Western blot - Anti-DUSP4 antibody [EPR19881] (ab216576) at 1/1000 dilution

Lane 1:
Wild-type A549 cell lysate at 20 µg

Lane 2:
DUSP4 knockout A549 cell lysate at 20 µg

Lane 2:
Western blot - Human DUSP4 knockout A549 cell line (ab273859)

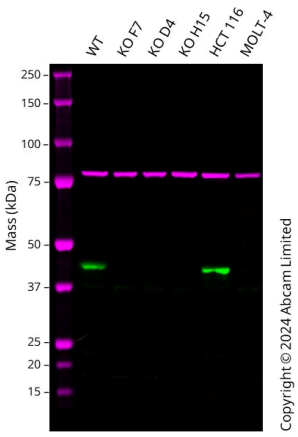
Lane 3:
A549 cell lysate at 20 µg

Lane 4:
MOLT-4 cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 43 kDa

Observed band size: 40 kDa



Western blot - Anti-DUSP4 antibody [EPR19881] - BSA and Azide free (ab222487)

DUSP4 Western blot staining using rabbit Anti-DUSP4 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab216576).

Western blot: Anti-DUSP4 antibody [EPR19881] (ab216576) staining at 1/1000 dilution, shown in green; Mouse anti-CANX [CANX/1543] (ab238078) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab216576 was shown to bind specifically to DUSP4. A band was observed at 40 kDa in wild-type A549 cell lysates with no signal observed at this size in DUSP4 knockout cell line. To generate this image, wild-type and DUSP4 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.

All lanes:
Western blot - Anti-DUSP4 antibody [EPR19881] (ab216576) at 1/1000 dilution

Lane 1:
Wild-type A549 cell lysate at 20 µg

Lanes 2 - 3:
DUSP4 knockout A549 cell lysate at 20 µg

Lane 4:
DUSP4 knockout A549 H15 cell lysate at 20 µg

Lane 5:
HCT 116 cell lysate at 20 µg

Lane 6:
MOLT-4 cell lysate at 20 µg

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.

Observed band size: 40 kDa

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