abcam

ab40809

Anti-ERK5 antibody [EP791Y]

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	EP791Y
Purification technique	Affinity purification Protein A
Concentration	0.514 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	1/30
Notes	**For unpurified use at 1/50 dilution.**

Expected

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

WB

Tested

Species	Human
Dilution info	1/10000
Notes	**For unpurified use at 1/1000 - 1/5000 dilution.**

Species	Mouse
Dilution info	1/10000
Notes	**For unpurified use at 1/1000 - 1/5000 dilution.**

Species	Rat
Dilution info	1/10000
Notes	**For unpurified use at 1/1000 - 1/5000 dilution.**

ICC/IF

Tested

Species	Human
Dilution info	1/100
Notes	**For unpurified use at 1/250 - 1/500 dilution.**

Expected

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

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/50
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. For unpurified use at 1/100 dilution.

Expected

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

Target data

See full target information MAPK7

Function	Plays a role in various cellular processes such as proliferation, differentiation and cell survival. The upstream activator of MAPK7 is the MAPK kinase MAP2K5. Upon activation, it translocates to the nucleus and phosphorylates various downstream targets including MEF2C. EGF activates MAPK7 through a Ras-independent and MAP2K5-dependent pathway. As part of the MAPK/ERK signaling pathway, acts as a negative regulator of apoptosis in cardiomyocytes via interaction with STUB1/CHIP and promotion of STUB1-mediated ubiquitination and degradation of ICER-type isoforms of CREM (By similarity). May have a role in muscle cell differentiation. May be important for endothelial function and maintenance of blood vessel integrity. MAP2K5 and MAPK7 interact specifically with one another and not with MEK1/ERK1 or MEK2/ERK2 pathways. Phosphorylates SGK1 at Ser-78 and this is required for growth factor-induced cell cycle progression. Involved in the regulation of p53/TP53 by disrupting the PML-MDM2 interaction.

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

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	ERK5 also known as big MAP kinase 1 (BMK1) is an enzyme that functions as a protein-serine/threonine kinase within the MAP kinase family. It has a molecular weight of approximately 120 kDa. ERK5 is expressed in various tissues including the brain heart and lungs with notable expression in endothelial cells. The protein plays a mechanical role in signal transduction processes which it achieves by transmitting signals from extracellular sources to the cellular nucleus facilitating the regulation of gene expression.
Biological function summary	ERK5 influences cellular functions through its role in promoting cell proliferation and differentiation. It serves as part of a signaling complex that includes the MAP kinase kinase 5 (MEK5). This interaction allows ERK5 to phosphorylate and subsequently activate target substrates such as transcription factors promoting cellular responses necessary for development and survival. ERK5 also plays a role in the response to oxidative stress which contributes to its impact on survival pathways and cellular stress responses.
Pathways	ERK5 acts within the MAPK signaling cascade and is linked to the ERK/MAPK pathway and the phosphoinositide 3-kinase (PI3K) pathway. Within these pathways ERK5 interacts with other MAP kinases and regulatory proteins such as Ras and Raf contributing to a wide range of cellular processes including growth migration and survival. Through these interactions ERK5 influences cellular dynamics and mediates responses to growth factors and stress stimuli.
Associated diseases and disorders	Abnormal activity of ERK5 has been implicated in various conditions including cardiovascular disease and cancer. Elevated ERK5 signaling can promote tumor progression and metastasis in certain cancers by enhancing tumor cell

proliferation and survival. In cardiovascular disorders ERK5 interacts with proteins like calcium/calmodulin-dependent protein kinase II (CaMKII) influencing pathological cardiac remodeling and hypertrophy. These associations highlight ERK5's impact on disease mechanisms and highlight its potential as a therapeutic target.

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

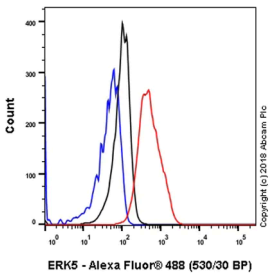
We do not recommend this combination. It is not covered by our product promise.

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.

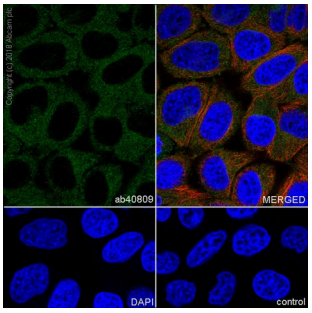
11 product images



Flow Cytometry (Intracellular) - Anti-ERK5 antibody [EP791Y] (ab40809)

ERK5 Flow Cytometry (Intracellular) staining using rabbit Anti-ERK5 antibody

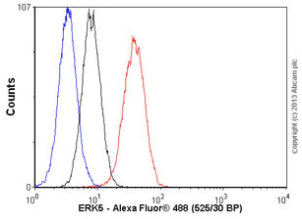
Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling ERK5 with purified ab40809 at 1/50 dilution (10 ug/ml) (red). Cells were fixed with 80% methanol. A Goat anti rabbit IgG (Alexa Fluorr®488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-ERK5 antibody [EP791Y] (ab40809)

ERK5 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-ERK5 antibody

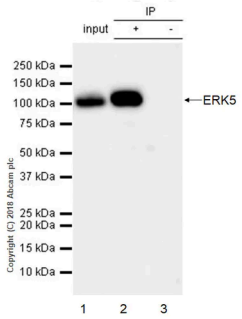
Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling ERK5 with Purified ab40809 at 1:100 (4.8 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Flow Cytometry (Intracellular) - Anti-ERK5 antibody [EP791Y] (ab40809)

ERK5 Flow Cytometry (Intracellular) staining using rabbit Anti-ERK5 antibody

Overlay histogram showing A549 cells stained with unpurified ab40809 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab40809, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

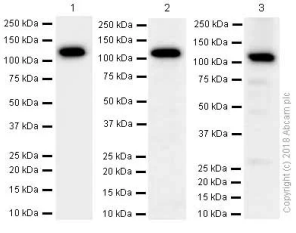


Immunoprecipitation - Anti-ERK5 antibody [EP791Y] (ab40809)

ab40809 (purified) at 1:30 dilution (2ug) immunoprecipitating ERK5 in HeLa whole cell lysate.
Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10ug
Lane 2 (+): ab40809 & HeLa whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab40809 in HeLa whole cell lysate
For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1:1000 dilution.
Blocking and diluting buffer: 5% NFDM/TBST.
All lanes:
Immunoprecipitation - Anti-ERK5 antibody [EP791Y] (ab40809)

Predicted band size: 88 kDa

Observed band size: 115 kDa



Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)

ERK5 Western blot staining using rabbit Anti-ERK5 antibody

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

All lanes:
Western blot - Anti-ERK5 antibody [EP791Y] (ab40809) at 1/2000 dilution

Lane 1:
HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates at 15 µg

Lane 2:
NIH/3T3 (Mouse embryonic fibroblast) whole cell lysates at 15 µg

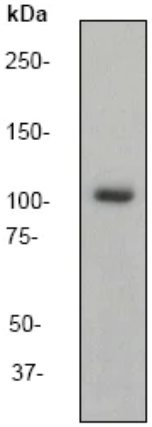
Lane 3:
PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates at 15 µg

Secondary

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

Predicted band size: 88 kDa

Observed band size: 115 kDa



Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)

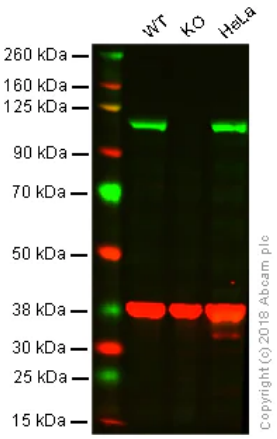
ERK5 Western blot staining of Hela cell lysate using rabbit Anti-ERK5 antibody

The predicted weight of 89 kDa is for the precursor version of human ERK5 protein. However, ab40809 detects endogenous levels of total Erk5 protein which appears around 115 kDa in SDS PAGE
All lanes:
Western blot - Anti-ERK5 antibody [EP791Y] (ab40809) at 1/5000 dilution

All lanes:
Hela cell lysate at 10 µg

Predicted band size: 88 kDa

Observed band size: 115 kDa



Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)

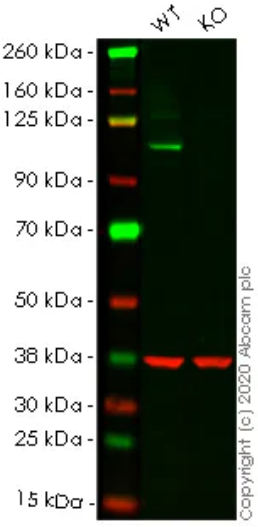
ERK5 Western blot staining using rabbit Anti-ERK5 antibody

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: MAPK7 (ERK5) knockout HAP1 whole cell lysate (20 µg)
Lane 3: HeLa whole cell lysate (20 µg)
Lanes 1 - 3: Merged signal (red and green). Green - ab40809 observed at 88 kDa. Red - loading control, ab9484, observed at 37 kDa.

Unpurified ab40809 was shown to specifically react with ERK5 in wild-type HAP1 cells as signal was lost in MAPK7 (ERK5) knockout cells. Wild-type and MAPK7 (ERK5) knockout samples were subjected to SDS-PAGE. Unpurified ab40809 and ab9484 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed 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 1/20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)

Predicted band size: 88 kDa



Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)

ERK5 Western blot staining using rabbit Anti-ERK5 antibody

Lanes 1- 2: Merged signal (red and green). Green - ab40809 observed at 115 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.
ab40809 was shown to react with ERK5 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265508 (knockout cell lysate ab258042) was used. Wild-type HeLa and MAPK7 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. ab40809 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) 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-ERK5 antibody [EP791Y] (ab40809) at 1/1000 dilution

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

Lane 2:
MAPK7 knockout HeLa cell lysate at 20 µg

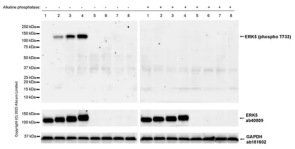
Lane 2:
Western blot - Human MAPK7 (ERK5) knockout HeLa cell line (ab265508)

Performed under reducing conditions.

Predicted band size: 88 kDa

Observed band size: 115 kDa

Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)



ERK5 Western blot staining using rabbit Anti-ERK5 antibody

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

In Western blot, ab323508 was shown to bind specifically to ERK5 (phospho T733). Target of interest was observed at 115 kDa in treated wild-type HeLa cell lysates (lanes -2-4) with no signal observed at this size in treated ERK5 knockout cell line (lanes 6-8) (lanes 5-8, knockout cell line ab265508 / knockout cell lysate ab258042).

Lane 1-8 on the left: untreated membrane;
Lane 1-8 on the right: membrane treated with alkaline phosphatase.

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

In Western blot, Anti-ERK5 antibody [EP791Y] - Total protein control (ab40809) staining at 1/1000 dilution.

All lanes:
Western blot - Anti-ERK5 (phospho T733) antibody [EPR28258-101] (ab323508) at 1/1000 dilution

Lane 1:
Untreated parental HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
Parental HeLa treated with 0.4M Sorbitol for 30min whole cell lysate at 20 µg

Lane 3:
Parental HeLa treated with 100nM Calyculin A for 30min whole cell lysate at 20 µg

Lane 4:
Parental HeLa treated with 0.4M Sorbitol and 100nM Calyculin A for 30min whole cell lysate at 20 µg

Lane 5:
Untreated ERK5 knockout HeLa whole cell lysate at 20 µg

Lane 6:
ERK5 knockout HeLa treated with 0.4M Sorbitol for 30min whole cell lysate at 20 µg

Lane 7:
ERK5 knockout HeLa treated with 100nM Calyculin A for 30min whole cell lysate at 20 µg

Lane 8:
ERK5 knockout HeLa treated with 0.4M Sorbitol and 100nM Calyculin A for 30min whole cell lysate at 20 µg

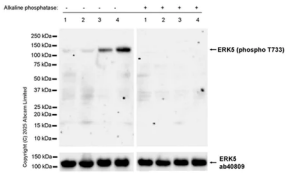
Secondary

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

Performed under reducing conditions.

Observed band size: 115 kDa, 36 kDa

Exposure time: 180s



Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)

ERK5 Western blot staining using rabbit Anti-ERK5 antibody

Lane 1-4 on the left: untreated membrane;
Lane 1-4 on the right: membrane treated with alkaline phosphatase.

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

In Western blot, Anti-ERK5 antibody [EP791Y] - Total protein control (ab40809) staining at 1/1000 dilution.

All lanes:
Western blot - Anti-ERK5 (phospho T733) antibody [EPR28258-101] (ab323508) at 1/1000 dilution

Lane 1:
Untreated NIH/3T3 (mouse embryonic fibroblast) whole cell lysate at 20 µg

Lane 2:
NIH/3T3 treated with 0.4M Sorbitol for 30min whole cell lysate at 20 µg

Lane 3:
NIH/3T3 treated with 100nM Calyculin A for 30min whole cell lysate at 20 µg

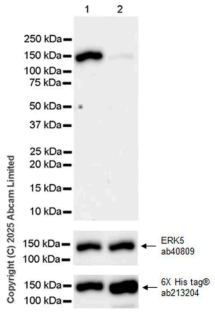
Lane 4:
NIH/3T3 treated with 0.4M Sorbitol and 100nM Calyculin A for 30min whole cell lysate at 20 µg

Secondary

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

Observed band size: 115 kDa

Exposure time: 180s



Western blot - Anti-ERK5 antibody [EP791Y] (ab40809)

ERK5 Western blot staining using rabbit Anti-ERK5 antibody

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

In Western blot, Anti-6X His tag® antibody [EPR20547] - ChIP Grade (ab213204) staining at 1/5000 dilution.

In Western blot, Anti-ERK5 antibody [EP791Y] - Total protein control (ab40809) staining at 1/1000 dilution.

All lanes:
Western blot - Anti-ERK5 (phospho T733) antibody [EPR28258-101] (ab323508) at 1/1000 dilution

Lane 1:
293T (human embryonic kidney epithelial cell) cells co-transfected with a human ERK5 WT expression vector containing a myc-His-tag® and a rat MEK5(S311D/T315D mutation)) expression vector containing a myc-His-tag®, whole cell lysate at 10 µg

Lane 2:
293T (human embryonic kidney epithelial cell) cells co-transfected with a human ERK5 (T733A mutation) expression vector containing a myc-His-tag® and a rat MEK5(S311D/T315D mutation)) expression vector containing a myc-His-tag®, whole cell lysate at 10 µg

Secondary

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

Observed band size: 115 kDa

Exposure time: 70s

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