abcam

Product datasheet

Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free ab218017

KO VALIDATED Recombinant RabMAb

3 References 9 Images

Overview

Product name	Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free
Description	Rabbit monoclonal [EPR17526] to ERK1 + ERK2 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC/IF, IP, WB
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa cell lysate. ICC/IF: HeLa cells. Flow Cyt (intra): A431 cells. IP. PC-12 whole cell extract.
General notes	ab218017 is the carrier-free version of **ab184699**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. 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. 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. 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**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR17526
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab218017 in the following tested applications.

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

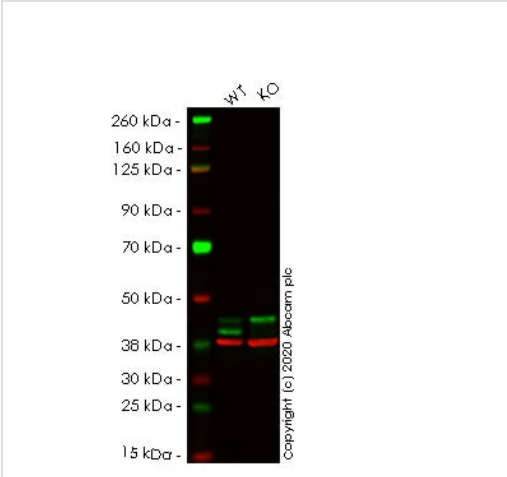
Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 44, 42 kDa (predicted molecular weight: 43, 41 kDa).

Target

Function	Involved in both the initiation and regulation of meiosis, mitosis, and postmitotic functions in differentiated cells by phosphorylating a number of transcription factors such as ELK1. Phosphorylates EIF4EBP1; required for initiation of translation. Phosphorylates microtubule-associated protein 2 (MAP2). Phosphorylates SPZ1 (By similarity). Phosphorylates heat shock factor protein 4 (HSF4) and ARHGEF2. Acts as a transcriptional repressor. Binds to a [GC]AAA[GC] consensus sequence. Repress the expression of interferon gamma-induced genes. Seems to bind to the promoter of CCL5, DMP1, IFIH1, IFITM1, IRF7, IRF9, LAMP3, OAS1, OAS2, OAS3 and STAT1. Transcriptional activity is independent of kinase activity.
Sequence similarities	Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. MAP kinase subfamily. Contains 1 protein kinase domain.
Domain	The TXY motif contains the threonine and tyrosine residues whose phosphorylation activates the MAP kinases.

Post-translational modifications	Dually phosphorylated on Thr-185 and Tyr-187, which activates the enzyme. Dephosphorylated by PTPRJ at Tyr-187.
Cellular localization	Nucleus.
Form	Mainly expressed in the cytoplasm and only localizes to the nucleus with treatment.

Images



Western blot - Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

All lanes : Anti-ERK1 + ERK2 antibody [EPR17526] (**ab184699**) at 1/10000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : MAPK1 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

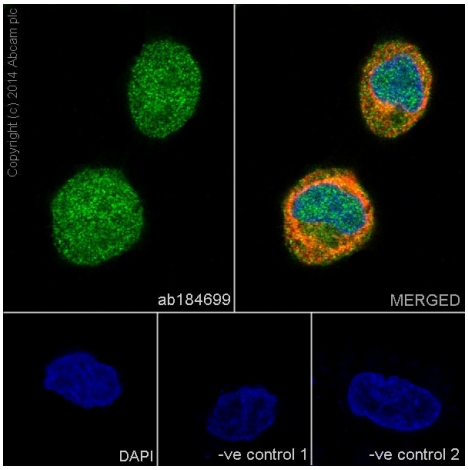
Performed under reducing conditions.

Predicted band size: 43, 41 kDa
Observed band size: 44 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab184699**).

Lanes 1-2: Merged signal (red and green). Green - **ab184699** observed at 44 kDa. Red - loading control **ab8245** observed at 37 kDa.

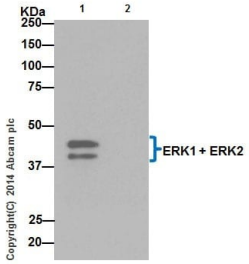
ab184699 Anti-ERK1 + ERK2 antibody [EPR17526] was shown to specifically react with ERK2 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265052** (knockout cell lysate **ab257525**) was used. Wild-type and ERK2 knockout samples were subjected to SDS-PAGE. **ab184699** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 10000 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-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling ERK1 + ERK2 with **ab184699** at 1/250 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/500 dilution (green). Confocal image showing both nuclear and cytoplasmic staining on HeLa cell line. The nuclear counter stain is DAPI (blue). Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows:
-ve control 1: **ab184699** at 1/250 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab184699**).



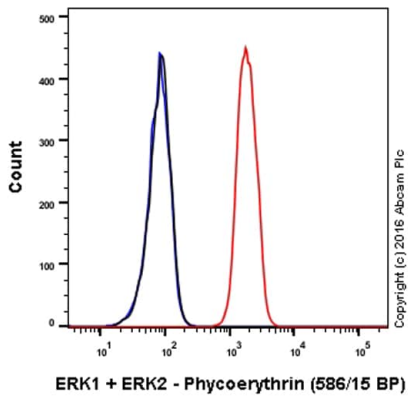
Immunoprecipitation - Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

ERK1 + ERK2 were immunoprecipitated from 1mg of PC-12 (Rat adrenal gland pheochromocytoma) whole cell extract with **ab184699** at 1/70 dilution. Western blot was performed from the immunoprecipitate using **ab184699** at 1/5000 dilution. Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG, was used as secondary antibody at 1/1500 dilution.

Lane 1: PC-12 whole cell extract. Lane 2: PBS instead of PC-12 whole cell extract.
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

44kDa band represents ERK1. 42kDa band represents ERK2.

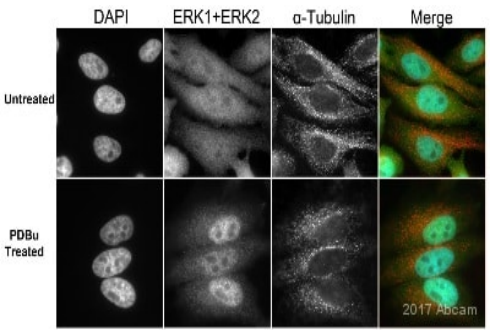
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab184699**).



Flow Cytometry (Intracellular) - Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

Clone EPR17526 (ab218017) has been successfully conjugated by Abcam. This image was generated using Anti-ERK1 + ERK2 antibody [EPR17526] (PE). Please refer to **ab212153** for protocol details.

Overlay histogram showing A431 cells stained with **ab212153** (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 90% methanol at -20°C for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (**ab212153**, 1/2500 dilution) for 30 min at 22°C.Isotype control antibody (black line) was rabbit IgG (monoclonal) Phycoerythrin (**ab209478**) used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 50 mW Yellow/Green laser (561nm) and 586/15 bandpass filter.

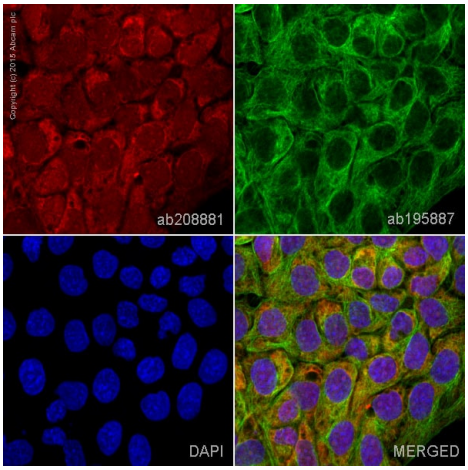


Immunocytochemistry/ Immunofluorescence - Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

This image is courtesy of an Abreview submitted by Kirk Mcmanus.

Ab184699 staining ERK1 + ERK2 in HeLa cells by ICC/IF (Immunocytochemistry/Immunofluorescence). Cells were fixed with paraformaldehyde and permeabilized with 0.5% Triton X-100. Samples were incubated with primary antibody (1/500 in PBS) for 1 hour at 22°C. A Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (**ab150081**) 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 (**ab184699**).

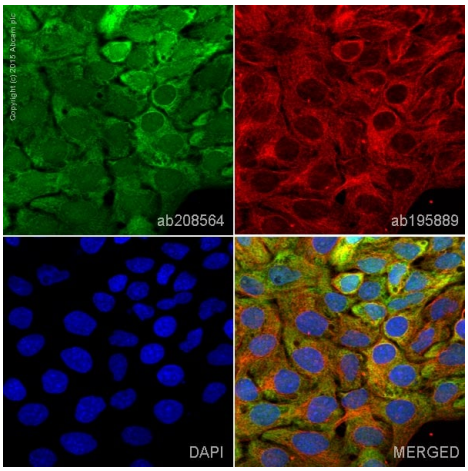


Immunocytochemistry/ Immunofluorescence - Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

Clone EPR17526 (ab218017) has been successfully conjugated by Abcam. This image was generated using Anti-ERK1 + ERK2 antibody [EPR17526] (Alexa Fluor® 647). Please refer to **ab208881** for protocol details.

ab208881 staining ERK1 + ERK2 in A431 cells. The cells were fixed with 100% methanol (5 min), 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 overnight at +4°C with **ab208881** at a 1/100 dilution (shown in red) and **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 488), at a 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



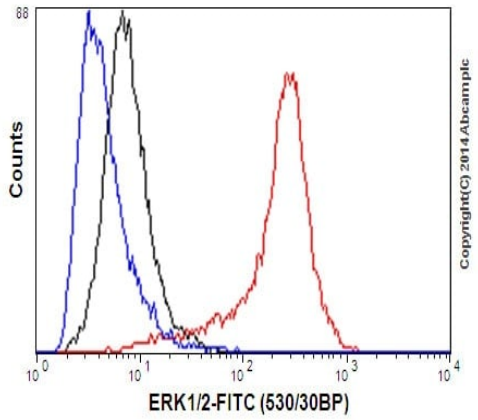
Immunocytochemistry/ Immunofluorescence - Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

Clone EPR17526 (ab218017) has been successfully conjugated by Abcam. This image was generated using Anti-ERK1 + ERK2 antibody [EPR17526] (Alexa Fluor® 488). Please refer to **ab208564** for protocol details.

ab208564 staining ERK1 + ERK2 in A431 cells. The cells were fixed with 100% methanol (5 min), 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 overnight at +4°C with **ab208564** at a 1/100 dilution (shown in green) and **ab195889**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at a 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

This product also gave a positive signal under the same testing conditions in A431 cells fixed with 4% formaldehyde (10 min).



Flow Cytometry (Intracellular) - Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

Intracellular flow cytometric analysis of A431 (Human epidermoid carcinoma) cells labeling ERK1 + ERK2 with **ab184699** at 1/440 dilution (red) compared with a rabbit monoclonal IgG isotype control (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (FITC) at 1/150 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 (**ab184699**).

Anti-ERK1 + ERK2 antibody [EPR17526] - BSA and Azide free (ab218017)

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