abcam

ab219584

Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] - BSA and Azide free

Recombinant RabMAb

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	EPR5693
Purification technique	Affinity purification Protein A
Specificity	This antibody will detect will detect JNK1 (pT183), JNK2 (pT183) and JNK3 (pT221).
Dissociation constant	2.09 x 10^{-11} M
Concentration	1.046 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

WB

Expected

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

Predicted

Species	Rat
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	(Heat to 98°C, allow to cool for 10-20 minutes)

Expected

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

Predicted

Species	Rat
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	-
Notes	-

Expected

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

Predicted

Species	Rat
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

Dot

Tested

Species	Synthetic peptide
Dilution info	-
Notes	-

Expected

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

Predicted

Species	Rat
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

Species	Human
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	ab199376 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

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

Predicted

Species	Rat
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

Target data

See full target information MAPK8 pT183

Function	Serine/threonine-protein kinase involved in various processes such as cell proliferation, differentiation, migration, transformation and programmed cell death. Extracellular stimuli such as pro-inflammatory cytokines or physical stress stimulate the stress-activated protein kinase/c-Jun N-terminal kinase (SAP/JNK) signaling pathway (PubMed:28943315). In this cascade, two dual specificity kinases MAP2K4/MKK4 and MAP2K7/MKK7 phosphorylate and activate MAPK8/JNK1. In turn, MAPK8/JNK1 phosphorylates a number of transcription factors, primarily components of AP-1 such as JUN, JDP2 and ATF2 and thus regulates AP-1 transcriptional activity (PubMed:18307971). Phosphorylates the replication licensing factor CDT1, inhibiting the interaction between CDT1 and the histone H4 acetylase HBO1 to replication origins (PubMed:21856198). Loss of this interaction abrogates the acetylation required for replication initiation (PubMed:21856198). Promotes stressed cell apoptosis by phosphorylating key regulatory factors including p53/TP53 and Yes-associates protein YAP1 (PubMed:21364637). In T-cells, MAPK8 and MAPK9 are required for polarized differentiation of T-helper cells into Th1 cells. Contributes to the survival of erythroid cells by phosphorylating the antagonist of cell death BAD upon EPO stimulation (PubMed:21095239). Mediates starvation-induced BCL2 phosphorylation, BCL2 dissociation from BECN1, and thus activation of autophagy (PubMed:18570871). Phosphorylates STMN2 and hence regulates microtubule dynamics, controlling neurite elongation in cortical neurons (By similarity). In the developing brain, through its cytoplasmic activity on STMN2, negatively regulates the rate of exit from multipolar stage and of radial migration from the ventricular zone (By similarity). Phosphorylates several other substrates including heat shock factor protein 4 (HSF4), the deacetylase SIRT1, ELK1, or the E3 ligase ITCH (PubMed:16581800, PubMed:17296730, PubMed:20027304). Phosphorylates the CLOCK-BMAL1 heterodimer and plays a role in the regulation of the circadian clock (PubMed:22441692). Phosphorylates the heat shock transcription factor HSF1, suppressing HSF1-induced transcriptional activity (PubMed:10747973). Phosphorylates POU5F1, which results in the inhibition of POU5F1's transcriptional activity and enhances its proteasomal degradation (By similarity). Phosphorylates JUND and this phosphorylation is inhibited in the presence of MEN1 (PubMed:22327296). In neurons, phosphorylates SYT4 which captures neuronal dense core vesicles at synapses (By similarity). Phosphorylates EIF4ENIF1/4-ET in response to oxidative stress, promoting P-body assembly (PubMed:22966201). Phosphorylates SIRT6 in response to oxidative stress, stimulating its mono-ADP-ribosyltransferase activity (PubMed:27568560). Phosphorylates NLRP3, promoting assembly of the NLRP3 inflammasome (PubMed:28943315). Phosphorylates ALKBH5 in response to reactive oxygen species (ROS), promoting ALKBH5 sumoylation and inactivation (PubMed:34048572). JNK1 isoforms display different binding patterns: beta-1 preferentially binds to c-Jun, whereas alpha-1, alpha-2, and beta-2 have a similar low level of binding to both c-Jun or ATF2. However, there is no correlation between binding and phosphorylation, which is achieved at about the same efficiency by all isoforms.

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

ab219584 is the carrier-free version of ab124956.

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.

Supplementary info

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

Activity summary	JNK1 JNK2 and JNK3 are c-Jun N-terminal kinases also known as stress-activated protein kinases (SAPKs). These kinases play significant roles in cellular stress responses. Commonly referred to by their shorter names JNK1 JNK2 and JNK3 they possess molecular weights of approximately 46 kDa to 55 kDa depending on their isoforms. These kinases are widely expressed across various tissues with JNK1 and JNK2 seen in most tissues whereas JNK3 shows more expression in neural tissues. Activation of JNKs occurs through phosphorylation at threonine 183 (T183) for JNK1 and JNK2 and threonine 221 (T221) for JNK3.
Biological function summary	C-Jun N-terminal kinases play important roles in mediating responses to stress stimuli including cytokines and ultraviolet irradiation. They are known to be part of the MAPK signaling complex and have direct involvement in the regulation of

	genes connected to apoptosis and cellular proliferation. JNK1 and JNK2 in particular have broad cellular roles and influence the activity of transcription factors such as c-Jun impacting gene expression significantly. JNK3 meanwhile contributes more to neuronal apoptosis given its expression pattern.
Pathways	JNK1 JNK2 and JNK3 actively engage in the MAPK signaling pathway connecting with several upstream and downstream proteins like MKK4 and MKK7 which serve as upstream kinases and c-Jun which acts downstream. The MAPK pathway is critical for translating extracellular signals into a wide range of cellular processes. The involvement of JNKs in this pathway highlights their contribution to balancing cell survival and death signals placing them alongside related proteins like ERK and p38MAPK within the signaling hierarchy.
Associated diseases and disorders	The activation and regulation of JNK1 JNK2 and JNK3 have essential implications in neurodegenerative diseases and cancer. JNK3 in particular is associated with neurodegenerative conditions such as Alzheimer's disease due to its role in neuronal stress-induced apoptosis. Similarly aberrant JNK1 and JNK2 activation connects to various cancers through their influence on genes governing cell cycle and apoptosis often in tandem with oncogenes or tumor suppressors like p53 and Bcl-2. These associations highlight the therapeutic potential of targeting these kinases in disease treatment strategies.

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

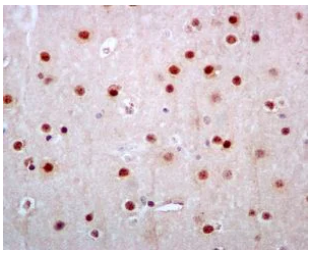
We do not recommend this combination. It is not covered by our product promise.

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.

6 product images

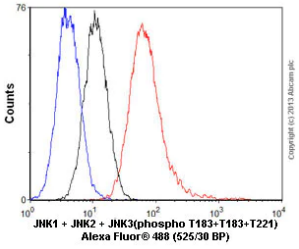


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] - BSA and Azide free (ab219584)

JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody

ab124956, at 1/100 dilution staining JNK1+JNK2+JNK3 in paraffin-embedded Human brain tissue, by Immunohistochemistry.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124956).

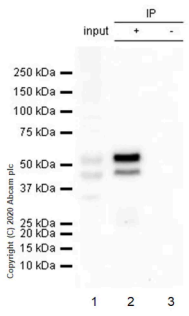
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] - BSA and Azide free (ab219584)

JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) Flow Cytometry (Intracellular) staining using rabbit Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody

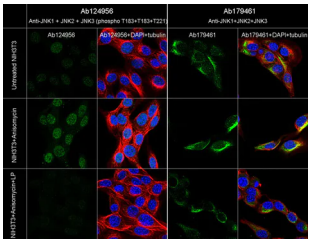
Overlay histogram showing HeLa cells stained with ab124956 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab124956, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was goat anti-rabbit Alexa Fluor® 488 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.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124956).



Immunoprecipitation - Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] - BSA and Azide free (ab219584)

This data was developed using ab124956, the same antibody clone in a different buffer formulation.
Purified ab124956 at 1/70 dilution (2µg) immunoprecipitating JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) in HeLa treated with 25ug/mL anisomycin for 30min whole cell lysate.
Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) treated with 25ug/mL anisomycin for 30min whole cell lysate 10µg
Lane 2 (+): ab124956 + HeLa treated with 25ug/mL anisomycin for 30min whole cell lysate.
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab124956 in HeLa treated with 25ug/mL anisomycin for 30min whole cell lysate.
VeriBlot for IP Detection Reagent (HRP) (ab131366) (1/5000 dilution) was used for Western

blotting.
Blocking Buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM/TBST.
Observed band size: 46, 54 kDa
All lanes:
Immunoprecipitation - Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] (ab124956)



Immunocytochemistry/ Immunofluorescence - Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] - BSA and Azide free (ab219584)

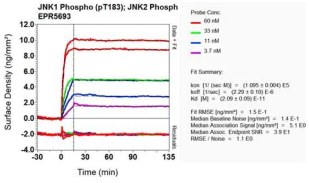
JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody

Immunocytochemistry/Immunofluorescence analysis of untreated, Anisomycin treated and Anisomycin + LP treated NIH/3T3 cells labelling JNK1 + JNK2 + JNK3 (phospho T183 + T183 + T221) with ab124956 at a dilution of 1/100 (left) and JNK1 + JNK2 + JNK3 with ab179461 at a dilution of 1/250 (right).
Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. ab7291, a mouse anti-tubulin (1/1000) and ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000) were also used.

The image shows increased nuclear staining after Anisomycin (250ng/ml, 30min) treatment on NIH3T3 cells. The LP treatment decreased the increased nuclear staining caused by Anisomycin.

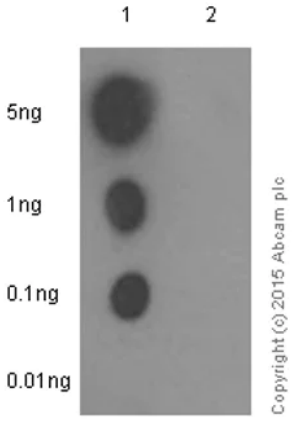
ab179461 was used as a Pan control for ab124956. The results showed cytoplasmic staining on untreated, Anisomycin and Anisomycin + LP treated NIH3T3 cells.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124956).



OI-RD Scanning - Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] - BSA and Azide free (ab219584)

We have systematically measured KD (the equilibrium dissociation constant between the antibody and its antigen), of more than 840 recombinant antibodies to assess not only their individual KD values but also to see the average affinity of antibody. Based on the comparison with published literature values for mouse monoclonal antibodies, Recombinant antibodies appear to be on average 1-2 order of magnitude higher affinity.



Dot Blot - Anti-JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) antibody [EPR5693] - BSA and Azide free (ab219584)

Dot blot analysis of JNK1/2/3 (pT183 + pT183 + pT221) peptide (Lane 1) and JNK1/2/3 non-phospho peptide (Lane 2) labelling JNK1 + JNK2 + JNK3 (phospho T183+T183+T221) with ab124956 at a dilution of 1/1000. ab97051 (Peroxidase conjugated goat anti-rabbit IgG (H+L)) was used as the secondary antibody at a dilution of 1/100000.
Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124956).

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