abcam

ab177030

Anti-ASK1 antibody [EP553Y] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EP553Y
Purification technique	Affinity purification Protein A
Concentration	1.029 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Expected

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

IP

Not recommended

Species	Mouse, Human
Dilution info	-
Notes	-

WB

Expected

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

ICC/IF

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

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

Target data

See full target information MAP3K5

Function	Serine/threonine kinase which acts as an essential component of the MAP kinase signal transduction pathway. Plays an important role in the cascades of cellular responses evoked by changes in the environment. Mediates signaling for determination of cell fate such as differentiation and survival. Plays a crucial role in the apoptosis signal transduction pathway through mitochondria-dependent caspase activation. MAP3K5/ASK1 is required for the innate immune response, which is essential for host defense against a wide range of pathogens. Mediates signal transduction of various stressors like oxidative stress as well as by receptor-mediated inflammatory signals, such as the tumor necrosis factor (TNF) or lipopolysaccharide (LPS). Once activated, acts as an upstream activator of the MKK/JNK signal transduction cascade and the p38 MAPK signal transduction cascade through the phosphorylation and activation of several MAP kinase kinases like MAP2K4/SEK1, MAP2K3/MKK3, MAP2K6/MKK6 and MAP2K7/MKK7. These MAP2Ks in turn activate p38 MAPKs and c-jun N-terminal kinases (JNKs). Both p38 MAPK and JNKs control the transcription factors activator protein-1 (AP-1).

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

ab177030 is the carrier-free version of ab45178.

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	The ASK1 protein also known as apoptosis signal-regulating kinase 1 is an important component in cellular stress response pathways. This 150 kDa protein is a member of the mitogen-activated protein kinase kinase kinase (MAP3K) family. ASK1 is expressed in several tissues including the heart brain liver and pancreas. Mechanically ASK1 gets activated in response to a variety of stress stimuli such as oxidative stress and endoplasmic reticulum stress which significantly impact the regulation of programmed cell death.
Biological function summary	In response to stressors ASK1 activates the c-Jun N-terminal kinase (JNK) and p38 MAPK signaling pathways which play roles in apoptosis and inflammation. ASK1 tends to form a complex with TRAF2 and TRAF6 key adapters in downstream signaling. This kinase acts as an important link bridging environmental stress signals to cellular responses. Through its function ASK1 influences cell fate decisions in response to various stress conditions.

Pathways	ASK1 is a central player in the MAPK signaling cascade. Within this cascade ASK1 regulates JNK and p38 MAPK pathways which are significant in stress adaptation and cellular responses. ASK1 interacts with proteins like MKK4 and MKK7 in the JNK pathway amplifying the stress signals. Through these interactions ASK1 ensures the propagation of signals important for adaptive responses or programmed cell death.
Associated diseases and disorders	Dysfunction in ASK1 activity associates with neurodegenerative diseases and cardiovascular disorders. In Alzheimer's disease for instance ASK1 contributes to neuronal apoptosis via its engagement with JNK signaling. In cardiovascular disorders ASK1 exacerbates heart failure by promoting cell death and inflammation. ASK1's interaction with proteins such as TNF receptor-associated factor 2 (TRAF2) highlights its role in these pathological conditions.

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

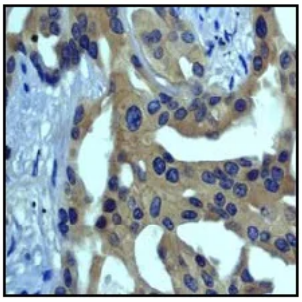
We do not recommend this combination. It is not covered by our product promise.

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

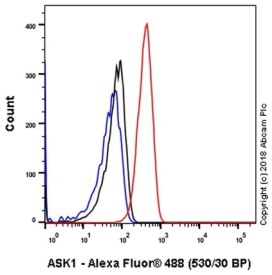


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ASK1 antibody [EP553Y] - BSA and Azide free (ab177030)

ASK1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ASK1 antibody

Unpurified ab45178 (1/10) staining human ASK1 in human lung carcinoma by immunohistochemistry using paraffin embedded tissue.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab45178).

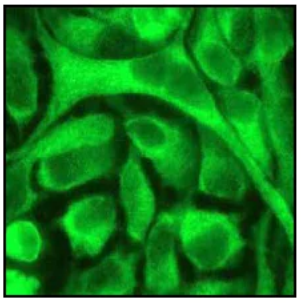
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-ASK1 antibody [EP553Y] - BSA and Azide free (ab177030)

ASK1 Flow Cytometry (Intracellular) staining using rabbit Anti-ASK1 antibody

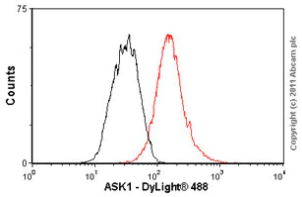
Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling ASK1 with purified ab45178 at 1/100 dilution (10 µg/ml) (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue). This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab45178).



Immunocytochemistry/ Immunofluorescence - Anti-ASK1 antibody [EP553Y] - BSA and Azide free (ab177030)

ASK1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-ASK1 antibody

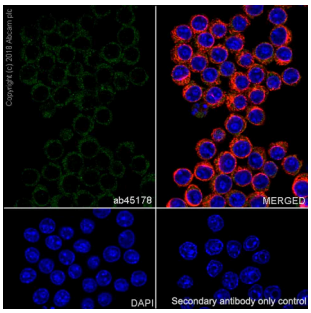
Immunofluorescent staining of HeLa cells using unpurified ab45178 (1/10).
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab45178).



Flow Cytometry (Intracellular) - Anti-ASK1 antibody [EP553Y] - BSA and Azide free (ab177030)

ASK1 Flow Cytometry (Intracellular) staining using rabbit Anti-ASK1 antibody

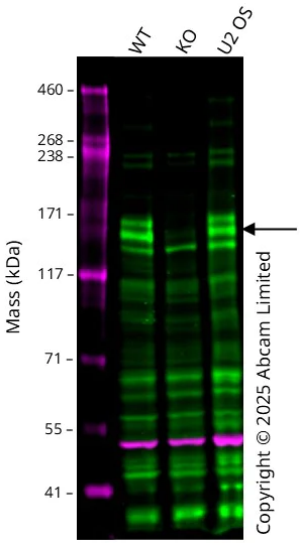
Overlay histogram showing HeLa cells stained with unpurified ab45178 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab45178, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was a goat anti-rabbit DyLight® 488 (IgG H+L) (ab96899) at 1/500 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. Acquisition of >5,000 events was performed.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab45178).



Immunocytochemistry/ Immunofluorescence - Anti-ASK1 antibody [EP553Y] - BSA and Azide free (ab177030)

ASK1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-ASK1 antibody

Immunocytochemistry/ Immunofluorescence analysis of RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) cells labeling ASK1 with Purified ab45178 at 1:100 (9.9 µg/ml). Cells were fixed in 100% Methanol. 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.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab45178).



Western blot - Anti-ASK1 antibody [EP553Y] - BSA and Azide free (ab177030)

ASK1 Western blot staining using rabbit Anti-ASK1 antibody

This data was developed using ab45178, the same antibody clone in a different buffer formulation.

Western blot: Rabbit Monoclonal[EP553Y] to ASK1 ab45178 staining at 1/2000 dilution, shown in green; Mouse anti alpha Tubulin (ab7291) loading control staining at 1/20,000 dilution, shown in magenta. A band was observed at 150-170 kDa in Wild-type A549 cell lysates with no signal observed at this size in MAP3K5 knockout A549 cell line. To generate this image, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3pc 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 800CW & Goat anti-Mouse 680RD at 1/20,000 dilution.
All lanes:
Western blot - Anti-ASK1 antibody [EP553Y] (ab45178) at 1/2000 dilution

Lane 1:
Wild-type A549 at 20 µg

Lane 2:
Western blot - Human MAP3K5 knockout A549 cell line (ab301060) at 20 µg

Lane 3:
U-2 OS at 20 µg

Secondary

Lanes 1 - 3:
Goat anti-Rabbit 800CW at 1/20000 dilution

Lanes 1 - 3:
Goat anti-Mouse 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 155 kDa

Observed band size: 150-170 kDa, 50 kDa

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