abcam

Product datasheet

Anti-MEK1 antibody [Y77] - BSA and Azide free ab167151





RabMAb

8 Images

Overview

Product name Anti-MEK1 antibody [Y77] - BSA and Azide free

Description Rabbit monoclonal [Y77] to MEK1 - BSA and Azide free

Host species Rabbit

Specificity The antibody does not crossreact with other MAP kinase kinase family members.

Tested applications Suitable for: Flow Cyt (Intra), WB, IP, ICC/IF, IHC-P

Species reactivity Reacts with: Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Wild-type HAP1 whole cell lysate; A431 and HepG2 whole cell lysates. IHC-P: Urinary

bladder carcinoma tissue. ICC/IF: HeLa cells.

General notes ab167151 is the carrier-free version of <u>ab32576</u>.

Our <u>carrier-free</u> 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 <u>conjugation kits</u> 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 Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal

Clone number Y77
Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab167151 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. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		Use at an assay dependent concentration. Detects a band of approximately 45 kDa (predicted molecular weight: 43 kDa).
IP		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.

Target

Function Catalyzes the concomitant phosphorylation of a threonine and a tyrosine residue in a Thr-Glu-Tyr

sequence located in MAP kinases. Activates ERK1 and ERK2 MAP kinases.

Tissue specificity Widely expressed, with extremely low levels in brain.

Involvement in disease Defects in MAP2K1 are a cause of cardiofaciocutaneous syndrome (CFC syndrome)

[MIM:115150]; also known as cardio-facio-cutaneous syndrome. CFC syndrome is characterized by a distinctive facial appearance, heart defects and mental retardation. Heart defects include pulmonic stenosis, atrial septal defects and hypertrophic cardiomyopathy. Some affected individuals present with ectodermal abnormalities such as sparse, friable hair, hyperkeratotic skin

lesions and a generalized ichthyosis-like condition. Typical facial features are similar to Noonan syndrome. They include high forehead with bitemporal constriction, hypoplastic supraorbital ridges, downslanting palpebral fissures, a depressed nasal bridge, and posteriorly angulated ears with prominent helices. The inheritance of CFC syndrome is autosomal dominant.

Sequence similarities

Belongs to the protein kinase superfamily. STE Ser/Thr protein kinase family. MAP kinase kinase subfamily.

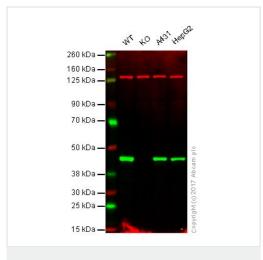
Contains 1 protein kinase domain.

Post-translational modifications

Phosphorylation on Ser/Thr by MAP kinase kinase kinases (RAF or MEKK1) regulates positively the kinase activity.

Acetylation by Yersinia yopJ prevents phosphorylation and activation, thus blocking the MAPK signaling pathway.

Images



Western blot - Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

Lane 2: MEK1 knockout HAP1 whole cell lysate (20 µg)

Lane 3: A431 whole cell lysate (20 µg)

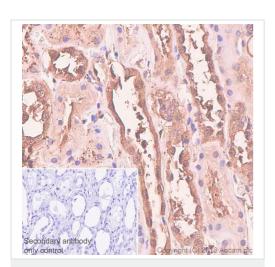
Lane 4: HepG2 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab32576</u> (unpurified) observed at 43 kDa. Red - loading control, <u>ab18058</u>, observed at 130 kDa.

ab32576 was shown to specifically react with MEK1 in wild-type HAP1 cells as signal was lost in MEK1 knockout cells. Wild-type and MEK1 knockout samples were subjected to SDS-PAGE.

Ab32576 and ab18058 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/10000 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.

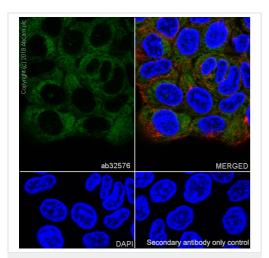
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32576).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human kidney tissue sections labeling MEK1 with purified ab32576 at 1/100 dilution (3.28 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

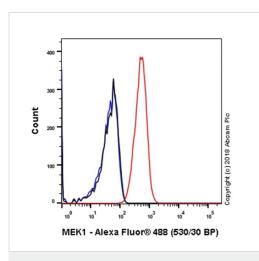
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32576).



Immunocytochemistry/ Immunofluorescence - Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

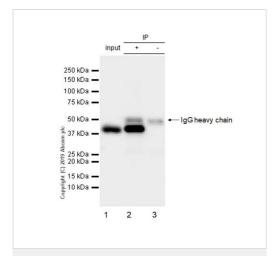
Immunocytochemistry/ Immunofluorescence analysis of A431 (Human epidermoid carcinoma epithelial cell) cells labeling MEK1 with purified $\underline{ab32576}$ at 1/50 dilution (6.6 µg/ml). Cells were fixed in 100% Methanol. Cells were counterstained with $\underline{ab195889}$ Antialpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1:200 (2.5 µg/ml). Goat anti rabbit lgG (Alexa Fluor® 488, $\underline{ab150077}$) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI (blue) 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 (<u>ab32576</u>).



Flow Cytometry (Intracellular) - Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling MEK1 with purified **ab32576** at 1/30 dilution (10µg/ml) (red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor[®] 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (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 (**ab32576**).



Immunoprecipitation - Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

<u>ab32576</u> (purified) at 1/20 dilution (2ug) immunoprecipitating
 MEK1 in Jurkat whole cell lysate. Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate 10ug

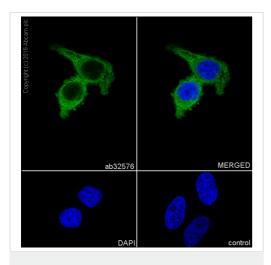
Lane 2 (+): ab32576 & Jurkat whole cell lysate

Lane 3 (-): Rabbit monoclonal $\lg G$ (ab172730) instead of ab32576 in Jurkat whole cell lysate

For western blotting, VeriBlot for IP secondary antibody (HRP) (ab131366) was used at 1/1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST.

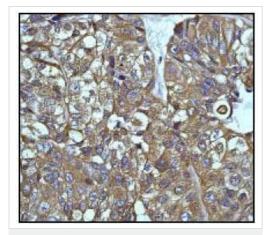
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32576).



Immunocytochemistry/ Immunofluorescence - Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

Immunocytochemistry/Immunofluorescence analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) labeling MEK1 with purified ab32576 at 1/500 dilution (5 µg/ml). Cells were fixed with 4% PFA and permeabilized with 0.1% triton X-100. ab150077 Goat anti rabbit IgG (Alexa Fluor[®] 488) at 1/1000 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI. PBS was used instead of the primary antibody as the negative control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32576).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

<u>ab32576</u> (unpurified) at a 1/250 dilution staining MEK1 in human urinary bladder carcinoma tissue by IHC-P.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32576).



Anti-MEK1 antibody [Y77] - BSA and Azide free (ab167151)

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