abcam

ab230795

Anti-TRAF2 antibody [EPR6048] - BSA and Azide free

Recombinant | RabMAb | KO Validated

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	EPR6048
Purification technique	Affinity purification Protein A
Dissociation constant	1.7 x 10^{-12} M
Concentration	1.019 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	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Mouse
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	-
Notes	-

Expected

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

Predicted

Species	Mouse
Dilution info	-
Notes	-

WB

Tested

Species	Rat
Dilution info	1/1000
Notes	-

Species	Human
Dilution info	-
Notes	-

Predicted

Species	Mouse
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Expected

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

Predicted

Species	Mouse
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	ab199376 - Rabbit monoclonal IgG (Low endotoxin, Azide free), is suitable for use as an isotype control with this antibody.

Expected

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

Predicted

Species	Mouse
Dilution info	-
Notes	-

Target data

See full target information TRAF2

Function

E3 ubiquitin-protein ligase that regulates activation of NF-kappa-B and JNK and plays a central role in the regulation of cell survival and apoptosis (PubMed:10346818, PubMed:11784851, PubMed:12917689, PubMed:15383523, PubMed:18981220, PubMed:19150425, PubMed:19810754, PubMed:19918265, PubMed:19937093, PubMed:20047764, PubMed:20064526, PubMed:20385093, PubMed:20577214, PubMed:22212761). Catalyzes 'Lys-63'-linked ubiquitination of target proteins, such as BIRC3, IKBKE, MLST8, RIPK1 and TICAM1 (PubMed:23453969, PubMed:28489822). Is an essential constituent of several E3 ubiquitin-protein ligase complexes, where it promotes the ubiquitination of target proteins by bringing them into contact with other E3 ubiquitin ligases (PubMed:15383523, PubMed:18981220). Regulates BIRC2 and BIRC3 protein levels by inhibiting their autoubiquitination and subsequent degradation; this does not depend on the TRAF2 RING-type zinc finger domain (PubMed:11907583, PubMed:19506082). Plays a role in mediating activation of NF-kappa-B by EIF2AK2/PKR (PubMed:15121867). In complex with BIRC2 or BIRC3, promotes ubiquitination of IKBKE (PubMed:23453969). Acts as a regulator of mTORC1 and mTORC2 assembly by mediating 'Lys-63'-linked ubiquitination of MLST8, thereby inhibiting formation of the mTORC2 complex, while facilitating assembly of the mTORC1 complex (PubMed:28489822). Required for normal antibody isotype switching from IgM to IgG (By similarity).

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

ab230795 is the carrier-free version of ab126758.

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	TRAF2 known also as TNF Receptor-Associated Factor 2 is an adapter protein weighing approximately 56 kDa. It is expressed in many tissues such as lymph nodes spleen and thymus. This protein acts as a signal transducer playing an integral role in the activation of NF-kB and MAPK signaling pathways. TRAF2 interacts with TNF receptors and other proteins using its RING finger and zinc finger domains for ubiquitination processes.
Biological function summary	TRAF2 modulates immune responses by regulating cell survival proliferation and apoptosis. It often forms complexes with other TRAF family proteins to mediate signal transmission. These complexes control the balance between pro-apoptotic and anti-apoptotic signals impacting the regulation of immune and inflammatory responses. Its interactions are necessary for the proper function of NF-kB ensuring balanced immune activities.
Pathways	TRAF2 is a critical component within both NF-kB and JNK signaling pathways. In the NF-kB pathway TRAF2 intersects with proteins like IKK and NEMO facilitating

	the transcription of genes involved in immune and inflammatory responses. In JNK signaling TRAF2 influences cellular responses through regulation of apoptotic signals. By linking these pathways TRAF2 ensures a coordinated cellular response to extracellular stimuli.
Associated diseases and disorders	TRAF2's dysregulation is linked with autoimmune diseases and certain types of cancer. Overactivity can contribute to chronic inflammation highlighting its role in autoimmune disorders like rheumatoid arthritis. In cancer TRAF2's involvement with TNF receptors can lead to uncontrolled cell proliferation. Proteins like cIAP1 and cIAP2 are often related to TRAF2 in these contexts influencing the progression and modulation of disease states.

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

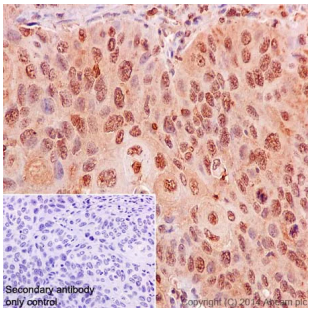
We do not recommend this combination. It is not covered by our product promise.

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.

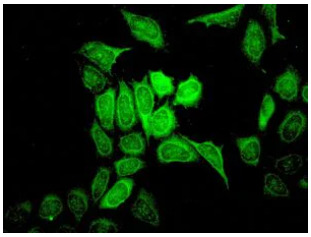
11 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

TRAF2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-TRAF2 antibody

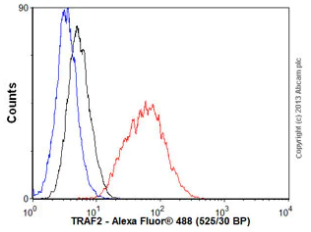
This IHC data was generated using the same anti-TRAF2 antibody clone, EPR6048, in a different buffer formulation (cat# ab126758).
Immunohistochemical staining of paraffin embedded human cervical cancer with purified ab126758 at a working dilution of 1/100. The secondary antibody used is ab97051, a HRP-conjugated goat anti-rabbit IgG (H+L), at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Immunocytochemistry/ Immunofluorescence - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

TRAF2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-TRAF2 antibody

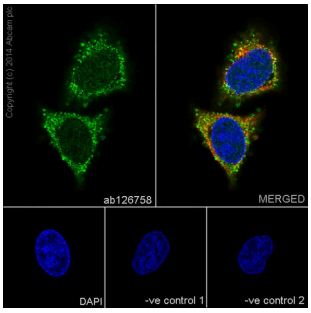
Unpurified ab126758, at 1/100 dilution, staining TRAF2 in HeLa cells, by Immunofluorescence. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab126758).



Flow Cytometry (Intracellular) - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

TRAF2 Flow Cytometry (Intracellular) staining using rabbit Anti-TRAF2 antibody

Overlay histogram showing HeLa cells stained with unpurifiedï¿½ab126758 (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 (unpurified ab126758, 1/100 dilution) for 30 min at 22ï¿½C. The secondary antibody used was Alexa Fluorï¿½ 488 goat anti-rabbit IgG (H+L) (ab150077) at 1/2000 dilution for 30 min at 22ï¿½C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1ï¿½g/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 (ab126758).

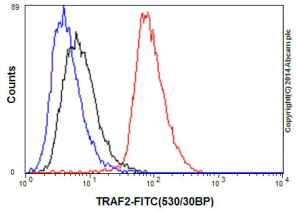


Immunocytochemistry/ Immunofluorescence - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

TRAF2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-TRAF2 antibody

Immunofluorescence staining of HeLa cells with purified ab126758 at a working dilution of 1 in 100, counter-stained with DAPI. Tubulin was stained with mouse anti-tubulin at a dilution of 1/1000 (ab7291) and Alexa Fluor® 594 goat anti-mouse at a dilution of 1/500 (ab150120) . The secondary antibody was ab150077 Alexa Fluor® 488 goat anti rabbit, used at a dilution of 1 in 500. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls

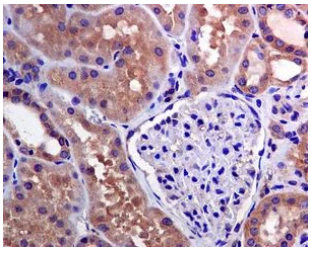
are shown in the bottom middle and right hand panels - for the first negative control, purified ab126758 was used at a dilution of 1/200 followed by an Alexa Fluor® 555 goat anti-mouse antibody at a dilution of 1/500 and for the second negative control mouse primary antibody (ab7291) and anti-rabbit secondary antibody (ab15007) were used.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab126758).



Flow Cytometry (Intracellular) - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

TRAF2 Flow Cytometry (Intracellular) staining using rabbit Anti-TRAF2 antibody

Overlay histogram showing HeLa cells fixed in 2% PFA and stained with purified ab126758 at a dilution of 1 in 120 (red line). The secondary antibody used was FITC goat anti-rabbit at a dilution of 1 in 150. Rabbit monoclonal IgG was used as an isotype control (black) and cells without antibody were used as a negative control (blue). This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab126758).

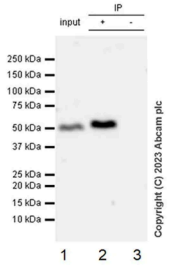


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

TRAF2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-TRAF2 antibody

Unpurified ab126758, at 1/50 dilution, staining TRAF2 in paraffin-embedded Human kidney tissue, by Immunohistochemistry.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab126758).

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



Immunoprecipitation - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

This data was developed using the same antibody clone in a different buffer formulation (ab126758). TRAF2 was immunoprecipitated from HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate with ab126758 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab126758 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution. Lane 1: HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate, 10µg Lane 2: HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab126758 in HeLa whole cell lysate Blocking and dilution buffer and concentration: 5% NFDM/TBST. Observed MV: 55kDa. Exposure time: 58 seconds.
All lanes:
Immunoprecipitation - Anti-TRAF2 antibody [EPR6048] (ab126758) at 1/1000 dilution

Lane 1:
HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate at 10 µg

Lane 2:
HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate

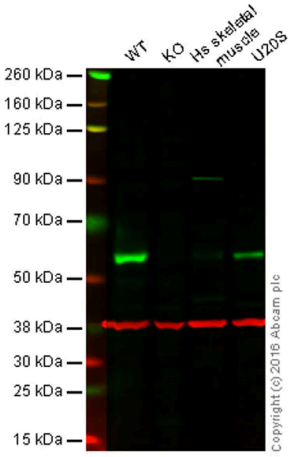
Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab126758 in HeLa whole cell lysate

Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Observed band size: 55 kDa

Exposure time: 58s



Western blot - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

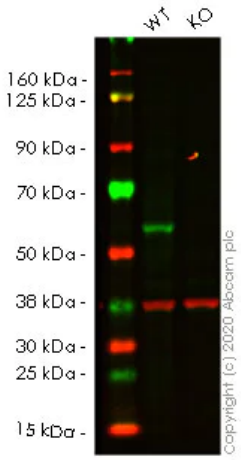
TRAF2 Western blot staining using rabbit Anti-TRAF2 antibody

This WB data was generated using the same anti-TRAF2 antibody clone, EPR6048, in a different buffer formulation (cat# ab126758).
Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: TRAF2 knockout HAP1 cell lysate (20 µg)
Lane 3: Human skeletal muscle lysate (20 µg)
Lane 4: U2OS cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab126758 observed at 55 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab126758 was shown to specifically react with TRAF2 when TRAF2 knockout samples were used. Wild-type and TRAF2 knockout samples were subjected to SDS-PAGE. ab126758 and ab8245 (loading control to GAPDH) were diluted 1/1000 and 1/2000 respectively and incubated overnight at 4°C. 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 1/10000 dilution for 1 h at room temperature before imaging.

All lanes:
Western blot - Anti-TRAF2 antibody [EPR6048] (ab126758)

Predicted band size: 55 kDa



Western blot - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

TRAF2 Western blot staining using rabbit Anti-TRAF2 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab126758).
Lanes 1- 2: Merged signal (red and green). Green - ab126758 observed at 55 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.

ab126758 was shown to react with TRAF2 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266060 (knockout cell lysate ab257759) was used. Wild-type HEK-293T and TRAF2 knockout HEK-293T 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. ab126758 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-TRAF2 antibody [EPR6048] (ab126758) at 1/1000 dilution

Lane 1:
Wild-type HEK-293T cell lysate at 20 µg

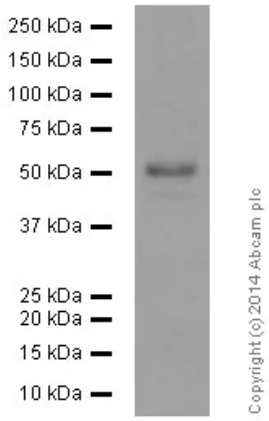
Lane 2:
TRAF2 knockout HEK-293T cell lysate at 20 µg

Lane 2:
Western blot - Human TRAF2 knockout HEK-293T cell line (ab266060)

Performed under reducing conditions.

Predicted band size: 55 kDa

Observed band size: 55 kDa



Western blot - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST
All lanes:
Western blot - Anti-TRAF2 antibody [EPR6048] (ab126758) at 1/1000 dilution

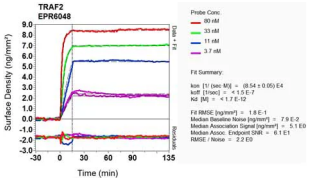
All lanes:
PC-12 whole cell lysate

Secondary

All lanes:
HRP Goat Anti-Rabbit (H+L) at 1/1000 dilution

Predicted band size: 55 kDa

Observed band size: 53 kDa



OI-RD Scanning - Anti-TRAF2 antibody [EPR6048] - BSA and Azide free (ab230795)

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.

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