abcam

ab197202

Anti-Caspase-3 antibody [E87] - 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	E87
Purification technique	Affinity purification Protein A
Concentration	1.022 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	**For unpurified, use 1/25 dilution.**

Not recommended

Species	Mouse
Dilution info	-
Notes	**For unpurified, use 1/25 dilution.**

Flow Cyt (Intra)

Tested

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

Not recommended

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

Target data

See full target information CASP3

Function Thiol protease that acts as a major effector caspase involved in the execution phase of apoptosis (PubMed:18723680, PubMed:20566630, PubMed:23650375, PubMed:35338844, PubMed:35446120, PubMed:7596430). Following cleavage and activation by initiator caspases (CASP8, CASP9 and/or CASP10), mediates execution of apoptosis by catalyzing cleavage of many proteins (PubMed:18723680, PubMed:20566630, PubMed:23650375, PubMed:7596430). At the onset of apoptosis, it proteolytically cleaves poly(ADP-ribose) polymerase PARP1 at a '216-Asp-|-Gly-217' bond (PubMed:10497198, PubMed:16374543, PubMed:7596430, PubMed:7774019). Cleaves and activates sterol regulatory element binding proteins (SREBPs) between the basic helix-loop-helix leucine zipper domain and

the membrane attachment domain (By similarity). Cleaves and activates caspase-6, -7 and -9 (CASP6, CASP7 and CASP9, respectively) (PubMed:7596430). Cleaves and inactivates interleukin-18 (IL18) (PubMed:37993714, PubMed:9334240). Involved in the cleavage of huntingtin (PubMed:8696339). Triggers cell adhesion in sympathetic neurons through RET cleavage (PubMed:21357690). Cleaves DSG2 in response to apoptosis resulting in a loss of full length DSG2 at desmosome cell junctions and subsequent loss of cell-cell adhesion (PubMed:17559062). Also cleaves JUP in response to apoptosis (PubMed:17559062). Cleaves and inhibits serine/threonine-protein kinase AKT1 in response to oxidative stress (PubMed:23152800). Acts as an inhibitor of type I interferon production during virus-induced apoptosis by mediating cleavage of antiviral proteins CGAS, IRF3 and MAVS, thereby preventing cytokine overproduction (PubMed:30878284). Also involved in pyroptosis by mediating cleavage and activation of gasdermin-E (GSDME) (PubMed:35338844, PubMed:35446120). Cleaves XRCC4 and phospholipid scramblase proteins XKR4, XKR8 and XKR9, leading to promote phosphatidylserine exposure on apoptotic cell surface (PubMed:23845944, PubMed:33725486). Cleaves BIRC6 following inhibition of BIRC6-caspase binding by DIABLO/SMAC (PubMed:36758104, PubMed:36758106).

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

ab197202 is the carrier-free version of ab32351.

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.

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.

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

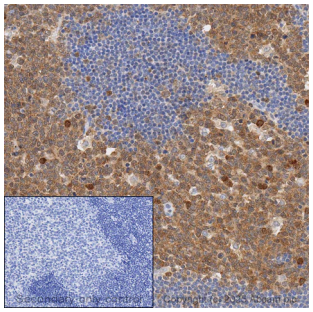
We do not recommend this combination. It is not covered by our product promise.

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.

9 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

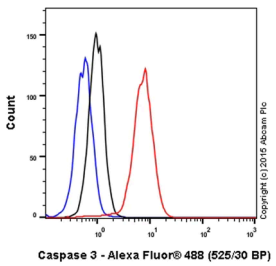
Caspase-3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human Tonsil tissue using rabbit Anti-Caspase-3 antibody

This data was developed using ab32351, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of formalin fixed paraffin embedded human tonsil labelling caspase-3 with ab32351 at a concentration of 0.1µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with a OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was performed with DISCOVERY cell conditioning solution (CC1) 100°C, pH8.5 for 32mins.

ab32351 anti-Caspase-3 antibody [E87] was incubated for 16mins at 37°C. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

Customers are encouraged to optimise antigen retrieval conditions, antibody concentration, incubation times and temperature for best results in their own IHC assay workflow (automated and manual)

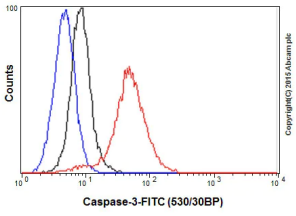


Flow Cytometry (Intracellular) - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

Caspase-3 Flow Cytometry (Intracellular) staining using rabbit Anti-Caspase-3 antibody

Overlay histogram showing HeLa cells stained with unpurified ab32351 (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 (ab32351, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluorr® 488 goat anti-rabbit IgG (H&L) (ab150081) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (ab172730, 0.1µg/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.

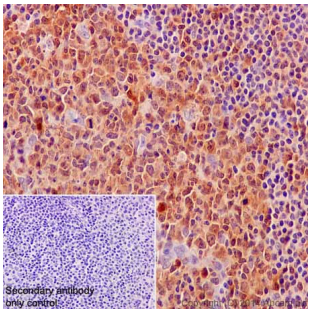
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32351).



Flow Cytometry (Intracellular) - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

Caspase-3 Flow Cytometry (Intracellular) staining using rabbit Anti-Caspase-3 antibody

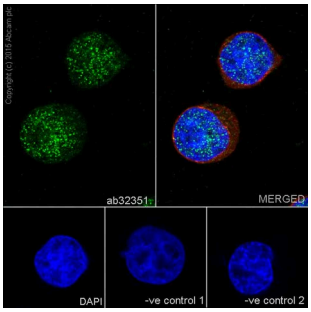
Overlay histogram showing Ramos cells fixed in 4% PFA and stained with purified ab32351 at a dilution of 1 in 180 (red line). The secondary antibody used was FITC goat anti-rabbit at a dilution of 1 in 500. Rabbit monoclonal IgG was used as an isotype control (black line) and cells incubated in the absence of both primary and secondary antibody were used as a negative control (blue line). This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32351).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

Caspase-3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Caspase-3 antibody

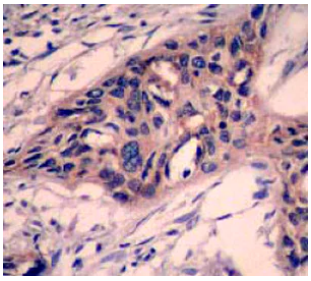
This IHC data was generated using the same anti-Caspase 3 antibody clone, E87, in a different buffer formulation (cat# ab32351).
Immunohistochemical staining of paraffin embedded human tonsil with purified ab32351 at a working dilution of 1/100. The secondary antibody used is HRP goat anti-rabbit IgG H&L (ab97051) at 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-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

Caspase-3 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Caspase-3 antibody

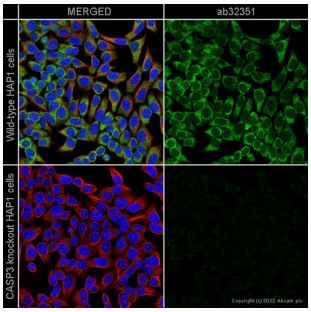
Immunofluorescence staining of Jurkat cells with purified ab32351 at a working dilution of 1/500, counter-stained with DAPI. The secondary antibody was Alexa Fluor® 488 goat anti-rabbit (ab150077), used at a dilution of 1/1000. ab7291, a mouse anti-tubulin antibody (1/1000), was used to stain tubulin along with ab150120 (Alexa Fluor® 594 goat anti-mouse, 1/1000), shown in the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified ab32351 was used at a dilution of 1/500 followed by an Alexa Fluor® 594 goat anti-mouse antibody (ab150120) at a dilution of 1/500. For negative control 2, ab7291 (mouse anti-tubulin) was used at a dilution of 1/500 followed by an Alexa Fluor® 488 goat anti-rabbit antibody (ab150077) at a dilution of 1/400.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32351).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

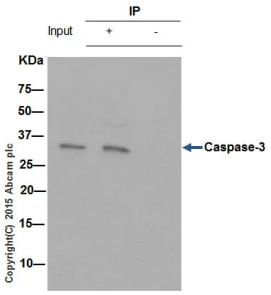
Caspase-3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Caspase-3 antibody

Unpurified ab32351, at a 1/25 dilution, staining Capase-3 in paraffin embedded human cervical carcinoma tissue by Immunohistochemistry.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32351).



Immunocytochemistry/ Immunofluorescence - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

This data was developed using the same antibody clone in a different buffer formulation (ab32351). ab32351 staining Caspase-3 in wild-type Hap1 cells (top panel) and CASP3 knockout Hap1 cells (bottom panel). The cells were fixed with 100% methanol (5 min) then 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 with ab32351 at 1µg/ml concentration and ab7291 (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor® 488) (ab150081) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor® 594) (ab150120) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI. Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).

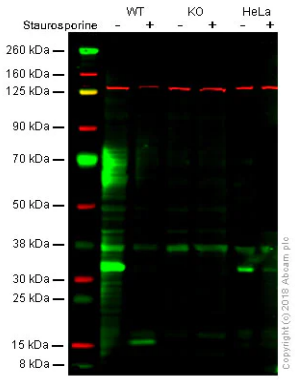


Immunoprecipitation - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

ab32351 (purified) at 1/50 immunoprecipitating Cullin 1 in 10 µg HeLa whole cell lysate (Lanes 1 and 2, observed at 35 kDa). Lane 3 - PBS. For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1500 dilution. Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32351).

All lanes:
Immunoprecipitation - Anti-Caspase-3 antibody [E87] (ab32351)

Predicted band size: 31 kDa



Western blot - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202)

Lanes 1 - 4: Merged signal (red and green). Green - ab32351 observed at 31 kDa. Red - loading control, ab130007, observed at 130 kDa.
ab32351 was shown to recognize Caspase 3 in wild-type HAP1 cells as signal was lost at the expected MW in HAP1 Staurosporine Treated (CASP3) knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and HAP1 Staurosporine Treated (CASP3) knockout samples were subjected to SDS-PAGE. ab32351 and ab130007 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/5000 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 (ab32351).

All lanes:
Western blot - Anti-Caspase-3 antibody [E87] - BSA and Azide free (ab197202) at 1/5000 dilution

Lane 1:
DMSO control wild-type HAP1 whole cell lysate at 20 µg

Lane 2:
Staurosporine treated wild-type HAP1 whole cell lysate at 20 µg

Lane 3:
DMSO control CASP3 knockout HAP1 whole cell lysate at 20 µg

Lane 4:
Staurosporine treated CASP3 knockout HAP1 whole cell lysate at 20 µg

Predicted band size: 31 kDa

Observed band size: 31 kDa

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