abcam

Product datasheet

Anti-Bid antibody [Y8] - BSA and Azide free ab247217

Recombinant RabMAb

6 Images

Overview

Product name	Anti-Bid antibody [Y8] - BSA and Azide free
Description	Rabbit monoclonal [Y8] to Bid - BSA and Azide free
Host species	Rabbit
Specificity	This antibody does not cross-react with other Bcl-2 family members.
Tested applications	**Suitable for:** IHC-P, Flow Cyt (Intra), IP, WB, ICC/IF
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	ICC/IF: MCF7 cells IP: Jurkat whole cell lysate.
General notes	ab247217 is the carrier-free version of **ab32060**. Our **carrier-free** 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 **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. 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**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	Y8
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab247217 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

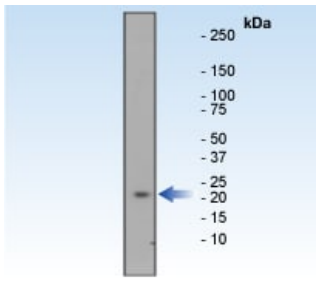
Application	Abreviews	Notes
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
Flow Cyt (Intra)		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 22 kDa (predicted molecular weight: 22 kDa).
ICC/IF		Use at an assay dependent concentration.

Target

Function	The major proteolytic product p15 BID allows the release of cytochrome c (By similarity). Isoform 1, isoform 2 and isoform 4 induce ICE-like proteases and apoptosis. Isoform 3 does not induce apoptosis. Counters the protective effect of Bcl-2.
Tissue specificity	Isoform 2 and isoform 3 are expressed in spleen, bone marrow, cerebral and cerebellar cortex. Isoform 2 is expressed in spleen, pancreas and placenta (at protein level). Isoform 3 is expressed in lung, pancreas and spleen (at protein level). Isoform 4 is expressed in lung and pancreas (at protein level).
Domain	Intact BH3 motif is required by BIK, BID, BAK, BAD and BAX for their pro-apoptotic activity and for their interaction with anti-apoptotic members of the Bcl-2 family.

Post-translational modifications	TNF-alpha induces a caspase-mediated cleavage of p22 BID into a major p15 and minor p13 and p11 products. Phosphorylated upon DNA damage, probably by ATM or ATR. p15 BID is ubiquitinated by ITCH; ubiquitination results in proteasome-dependent degradation.
Cellular localization	Cytoplasm; Cytoplasm. Mitochondrion membrane. When uncleaved, it is predominantly cytoplasmic; Mitochondrion membrane. A significant proportion of isoform 2 localizes to mitochondria, it may be cleaved constitutively; Mitochondrion membrane. Associated with the mitochondrial membrane and Mitochondrion membrane. Translocates to mitochondria as an integral membrane protein.

Images

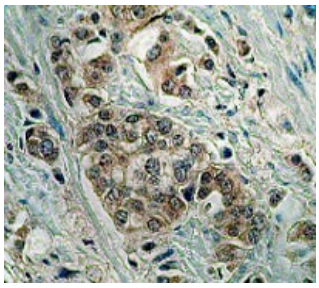


Western blot - Anti-Bid antibody [Y8] - BSA and Azide free (ab247217)

Anti-Bid antibody [Y8] (**ab32060**) at 1/1000 dilution + Jurkat cell lysate

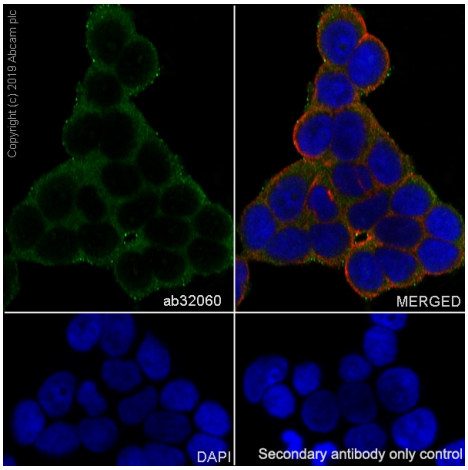
Predicted band size: 22 kDa
Observed band size: 22 kDa

This data was developed using **ab32060**, the same antibody clone in a different buffer formulation.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bid antibody [Y8] - BSA and Azide free (ab247217)

This data was developed using **ab32060**, the same antibody clone in a different buffer formulation.Immunohistochemical analysis of paraffin-embedded human prostate carcinoma using **ab32060** at 1/100 dilution. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

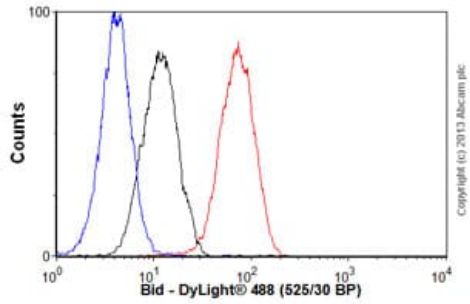


Immunocytochemistry/ Immunofluorescence - Anti-Bid antibody [Y8] - BSA and Azide free (ab247217)

This data was developed using the same antibody clone in a different buffer formulation (**ab32060**).
Immunocytochemistry analysis of MCF7 (human breast adenocarcinoma epithelial cell) labeling Bid with purified **ab32060** at 1/500 dilution (10 µg/ml). Cells were fixed with 100% methanol. Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/1000 (2 µg/ml) was used as the secondary antibody. **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 µg/ml) was used as counterstain. Nuclei were stained blue with DAPI.
Negative control: PBS instead of the primary antibody.

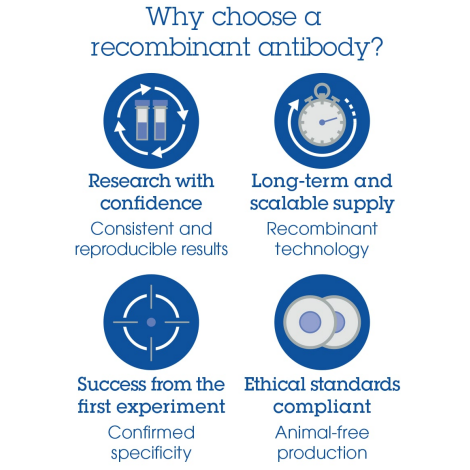
Immunoprecipitation - Anti-Bid antibody [Y8] - BSA and Azide free (ab247217)

This data was developed using **ab32060**, the same antibody clone in a different buffer formulation.
Purified **ab32060** at 1/50 dilution (2µg) immunoprecipitating Bid in Jurkat whole cell lysate.
Lane 1 (input): Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate 10µg
Lane 2 (+): **ab32060** + Jurkat whole cell lysate.
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab32060** in Jurkat whole cell lysate.
VeriBlot for IP Detection Reagent (HRP) (**ab131366**) (1/1000 dilution) was used for Western blotting.
Blocking Buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM/TBST.
Observed band size: 22 kDa



Flow Cytometry (Intracellular) - Anti-Bid antibody [Y8] - BSA and Azide free (ab247217)

This data was developed using **ab32060**, the same antibody clone in a different buffer formulation.Overlay histogram showing HeLa cells stained with **ab32060** (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 (**ab32060**, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit 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. Unlabelled sample (blue line). Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



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