abcam

Product datasheet

Anti-Bax antibody [EPR18284] ab182734

KO VALIDATED Recombinant RabMAb

51 References 9 Images

Overview

Product name	Anti-Bax antibody [EPR18284]
Description	Rabbit monoclonal [EPR18284] to Bax
Host species	Rabbit
Specificity	Expression levels of BAX protein vary with sample type. Induction may be required if endogenous expression is low.
Tested applications	**Suitable for:** WB, IP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa, MCF7, HuT-78, Ramos, HEK-293, Raji, L-929, C2C12, C6, PC-12 and NIH/3T3 whole cell lysates; Human kidney and spleen lysates; Mouse brain, heart, kidney and spleen lysates; Rat brain, kidney and spleen lysates; IP: HeLa whole cell lysate.
General notes	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 short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal

Clone number	EPR18284
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab182734 in the following tested applications.

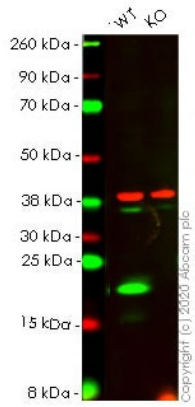
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/1000. Detects a band of approximately 19 kDa (predicted molecular weight: 21 kDa).
IP		1/40.

Target

Function	Accelerates programmed cell death by binding to, and antagonizing the apoptosis repressor BCL2 or its adenovirus homolog E1B 19k protein. Under stress conditions, undergoes a conformation change that causes translocation to the mitochondrion membrane, leading to the release of cytochrome c that then triggers apoptosis. Promotes activation of CASP3, and thereby apoptosis.
Tissue specificity	Expressed in a wide variety of tissues. Isoform Psi is found in glial tumors. Isoform Alpha is expressed in spleen, breast, ovary, testis, colon and brain, and at low levels in skin and lung. Isoform Sigma is expressed in spleen, breast, ovary, testis, lung, colon, brain and at low levels in skin. Isoform Alpha and isoform Sigma are expressed in pro-myelocytic leukemia, histiocytic lymphoma, Burkitt's lymphoma, T-cell lymphoma, lymphoblastic leukemia, breast adenocarcinoma, ovary adenocarcinoma, prostate carcinoma, prostate adenocarcinoma, lung carcinoma, epidermoid carcinoma, small cell lung carcinoma and colon adenocarcinoma cell lines.
Sequence similarities	Belongs to the Bcl-2 family.
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.
Cellular localization	Cytoplasm and Mitochondrion membrane. Cytoplasm. Colocalizes with 14-3-3 proteins in the cytoplasm. Under stress conditions, undergoes a conformation change that causes release from JNK-phosphorylated 14-3-3 proteins and translocation to the mitochondrion membrane.

Images



Western blot - Anti-Bax antibody [EPR18284] (ab182734)

All lanes : Anti-Bax antibody [EPR18284] (ab182734) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : BAX knockout HeLa cell lysate

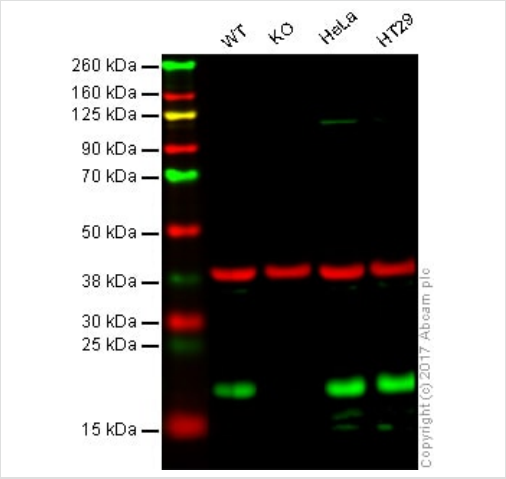
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 21 kDa
Observed band size: 21 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab182734 observed at 21 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab182734 was shown to react with Bax in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab255363** (knockout cell lysate **ab263841**) was used. Wild-type HeLa and BAX knockout HeLa 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. ab182734 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.



Western blot - Anti-Bax antibody [EPR18284] (ab182734)

All lanes : Anti-Bax antibody [EPR18284] (ab182734) at 1/1000 dilution

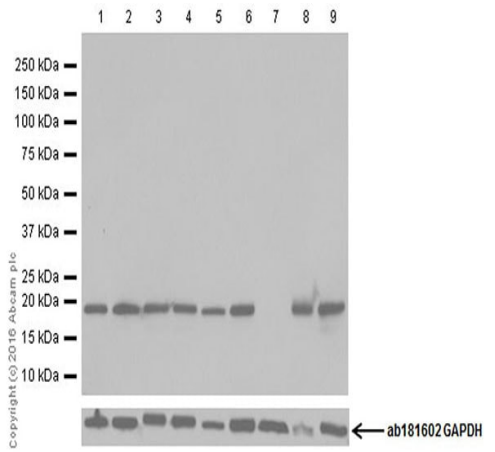
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : BAX knockout HAP1 whole cell lysate
Lane 3 : HeLa whole cell lysate
Lane 4 : HT29 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 21 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab182734 observed at 20 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab182734 was shown to specifically react with BAX when BAX knockout samples were used. Wild-type and BAX knockout samples were subjected to SDS-PAGE. Ab182734 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10000 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 1/10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Bax antibody [EPR18284] (ab182734)

All lanes : Anti-Bax antibody [EPR18284] (ab182734) at 1/1000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate
Lane 3 : HuT-78 (Human Sezary syndrome cell line) whole cell lysate
Lane 4 : Ramos (Human Burkitt's lymphoma cell line) whole cell lysate
Lane 5 : HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate
Lane 6 : Raji (Human Burkitt's lymphoma cell line) whole cell lysate
Lane 7 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate
Lane 8 : L-929 (Mouse connective tissue fibroblast cell line) whole cell lysate
Lane 9 : C2C12 (Mouse myoblast cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

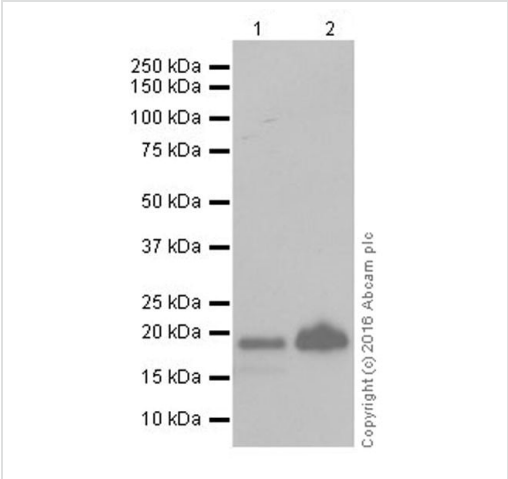
Predicted band size: 21 kDa
Observed band size: 19 kDa

Exposure time: 2 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

Negative control:

Jurkat doesn't express Bax. (PMID: 9583678).



Western blot - Anti-Bax antibody [EPR18284] (ab182734)

All lanes : Anti-Bax antibody [EPR18284] (ab182734) at 1/1000 dilution

Lane 1 : Human kidney lysate
Lane 2 : Human spleen lysate

Lysates/proteins at 10 µg per lane.

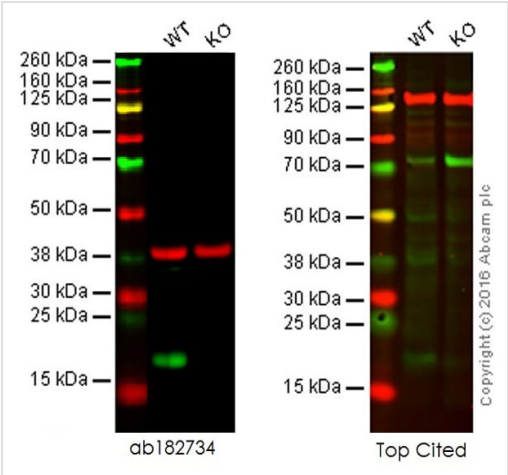
Secondary
All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 21 kDa
Observed band size: 19 kDa

Exposure time: 3 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

This product reacts with a 15 kD isoform (PMID: 7607685).



Western blot - Anti-Bax antibody [EPR18284] (ab182734)

All lanes : Anti-Bax antibody [EPR18284] (ab182734)

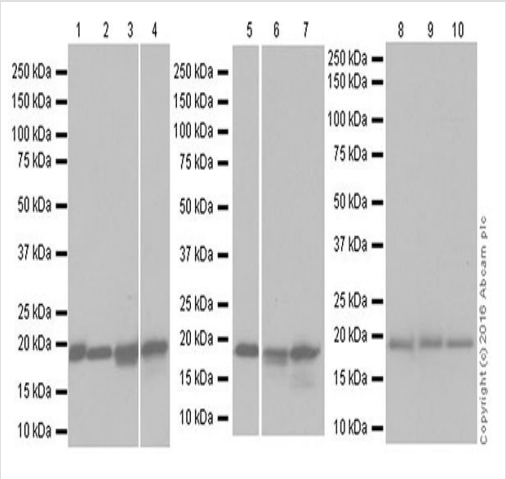
Lane 1 : Wild-type HAP1 cell lysate
Lane 2 : Bax knockout HAP1 cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 21 kDa

Lanes 1 - 2: Merged signal (red and green). Green - ab182734 observed at 20 kDa. Red - loading control, **ab8245**, observed at 37 kDa or **ab18058**, observed at 130 kDa.

This western blot image is a comparison between ab182734 and a competitor's top cited rabbit polyclonal antibody.



Western blot - Anti-Bax antibody [EPR18284] (ab182734)

All lanes : Anti-Bax antibody [EPR18284] (ab182734) at 1/1000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse heart lysate
Lane 3 : Mouse kidney lysate
Lane 4 : Mouse spleen lysate
Lane 5 : Rat brain lysate
Lane 6 : Rat kidney lysate
Lane 7 : Rat spleen lysate
Lane 8 : C6 (Rat glial tumor cell line) whole cell lysate
Lane 9 : PC-12 (Rat adrenal gland pheochromocytoma cell line) whole cell lysate
Lane 10 : NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

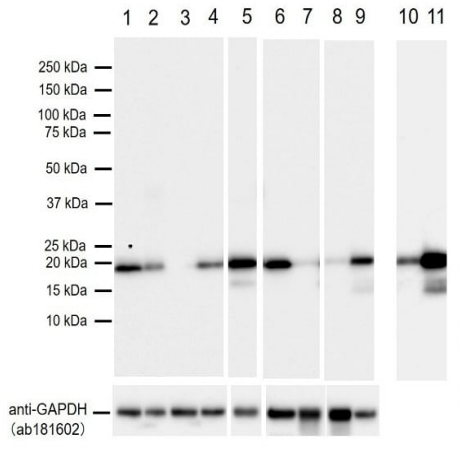
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 21 kDa
Observed band size: 19 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure times: Lane 1, 2 and 3: 3 seconds; Lane 4: 2 seconds; Lane 5: 3 seconds; Lane 6 and 7: 2 seconds; Lane 8, 9 and 10: 1 second.

This product reacts with a 15 kD isoform (PMID: 7607685).



Western blot - Anti-Bax antibody [EPR18284] (ab182734)

All lanes : Anti-Bax antibody [EPR18284] (ab182734) at 1/2000 dilution

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell). Whole cell lysates
Lane 2 : Hep G2 (Human hepatocellular carcinoma epithelial cell). Whole cell lysates
Lane 3 : Jurkat (Human T cell leukemia T lymphocyte) Whole cell lysates
Lane 4 : A549 (Human lung carcinoma epithelial cell) Whole cell lysates
Lane 5 : C2C12 (Mouse myoblasts myoblast) Whole cell lysates
Lane 6 : C6 (Rat glial tumor glial cell) Whole cell lysates
Lane 7 : Mouse brain. Whole tissue lysate
Lanes 8 & 10 : Rat brain. Whole tissue lysate
Lanes 9 & 11 : Rat Spleen. Whole tissue lysate

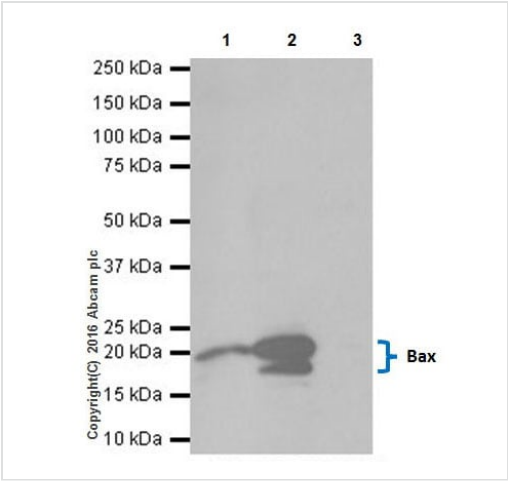
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

Predicted band size: 21 kDa
Observed band size: 18 kDa

Blocking and diluting 5% NFDM/TBST

Exposure time 1~9 lanes 24 s; 10~11 lanes 3 min



Immunoprecipitation - Anti-Bax antibody [EPR18284] (ab182734)

Bax was immunoprecipitated from 0.35 mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab182734 at 1/40 dilution. Western blot was performed from the immunoprecipitate using ab182734 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

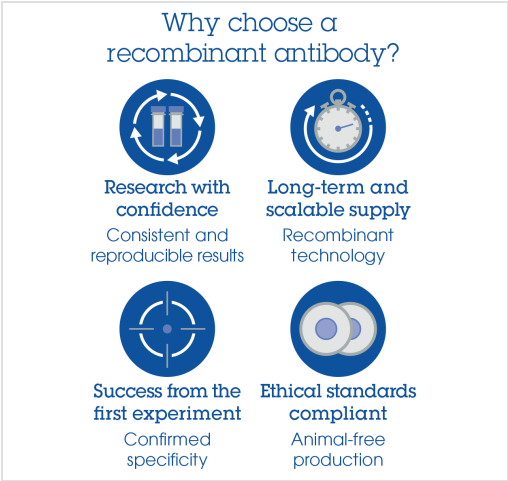
Lane 1: HeLa whole cell lysate, 10µg (Input).

Lane 2: ab182734 IP in HeLa whole cell lysate.

Lane 3: Rabbit IgG,monoclonal [EPR25A] - Isotype Control (**ab172730**) instead of ab182734 in HeLa whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 10 seconds.



Anti-Bax antibody [EPR18284] (ab182734)

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