abcam

Product datasheet

# Anti-Bax antibody [E63] - BSA and Azide free ab216985

敲除验证 重组 RabMAb

**3 References** 9 图像

## 概述

| | |
|---|---|
| **产品名称** | Anti-Bax抗体[E63] - BSA and Azide free |
| **描述** | 兔单克隆抗体[E63] to Bax - BSA and Azide free |
| **宿主** | Rabbit |
| **特异性** | Expression levels of BAX protein vary with sample type. Induction may be required if endogenous expression is low. |
| **经测试应用** | **适用于:** IHC-P, Sandwich ELISA, IP, WB<br>**不适用于:** Flow Cyt or ICC/IF |
| **种属反应性** | **与反应:** Mouse, Rat, Human<br>**预测可用于:** Cow |
| **免疫原** | Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. |
| **阳性对照** | WB: Recombinant Human Bax protein (Tagged) (**ab85157**), HeLa, HepG2, A549, C2C12 and C6 cell lysate. Wild-type Hap1 cell lysate. Rat spleen tissue lysate. IHC-P: Human lymph node and rat kidney tissues. Human lung carcinoma tissue. IP: HeLa cell lysate. |
| **常规说明** | ab216985 is the carrier-free version of **ab32503**.<br><br>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.<br><br>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.<br><br>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.<br><br>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.<br><br>This product is a recombinant monoclonal antibody, which offers several advantages including:<br>- High batch-to-batch consistency and reproducibility<br>- Improved sensitivity and specificity<br>- Long-term security of supply<br>- 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**.

## 性能

| | |
|---|---|
| **形式** | Liquid |
| **存放说明** | Shipped at 4°C. Store at +4°C. Do Not Freeze. |
| **存储溶液** | pH: 7.20<br>Constituent: PBS |
| **无载体** | 是 |
| **纯度** | Protein A purified |
| **克隆** | 单克隆 |
| **克隆编号** | E63 |
| **同种型** | IgG |

## 应用

**The Abpromise guarantee** **Abpromise™承诺保证使用ab216985于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

| 应用 | Ab评论 | 说明 |
|---|---|---|
| **IHC-P** | | Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. |
| **Sandwich ELISA** | | 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 21 kDa (predicted molecular weight: 21 kDa). |

**应用说明** Is unsuitable for Flow Cyt or ICC/IF.

## 靶标

**功能** 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.

**组织特异性** 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

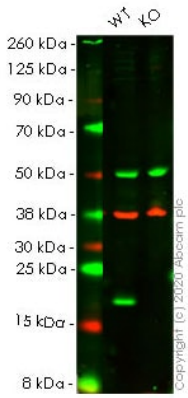
skin. Isoform Alpha and isoform Sigma are expressed in pro-myelocytic leukemia, histiocytic lymphoma, Burkitt's lymphoma, T-cell lymphoma, lymphoblastic leukemia, breast adenocarcinoma, ovary adenocarcinoma, prostate carcinoma, prostate adenocarcinoma, lung carcinoma, epidermoid carcinoma, small cell lung carcinoma and colon adenocarcinoma cell lines.

**序列相似性** Belongs to the Bcl-2 family.

**结构域** 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.

**细胞定位** 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.

## 图片




Western blot - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

**All lanes :** Anti-Bax antibody [E63] (**ab32503**) 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.

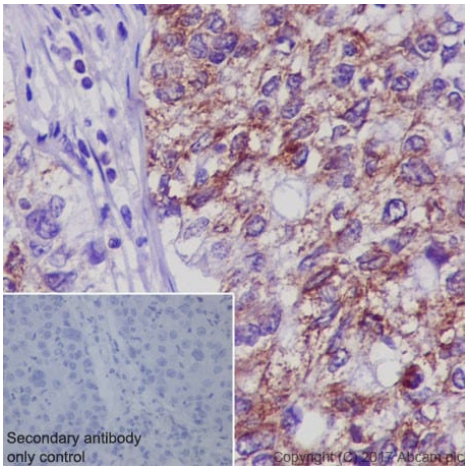
**Predicted band size:** 21 kDa
**Observed band size:** 21 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab32503**).

**Lanes 1- 2:** Merged signal (red and green). Green - **ab32503** observed at 21 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

**ab32503** 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. **ab32503** 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

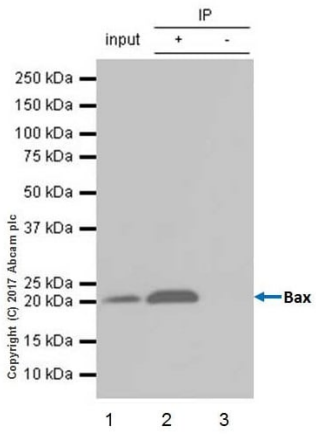
1 in 20000 dilution for 1 hour at room temperature before imaging.




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

Purified **ab32503** staining Bax in Human lung carcinoma tissue section by immmunohistochemistry (IHC-P- Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with paraffin and heat mediated antigen retrieval was performed using EDTA buffer (pH 9.0). Samples were incubated with primary antibody at 1:500 dilution. A goat anti-rabbit IgG H&L (HRP) (**ab97051**) was used as a secondary antibody at 1:500 dilution. Cytoplasmic staining on human lung carcinoma.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32503**).

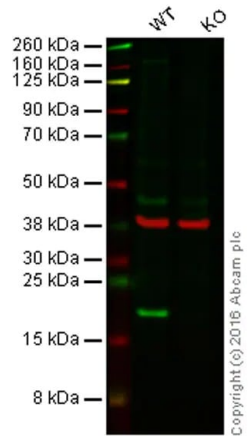



Immunoprecipitation - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

**Lane 1 (input):** HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate, 10µg
**Lane 2 (+):** HeLa whole cell lysate
**Lane 3 (-):** Rabbit monoclonal IgG (**ab172730**) instead of **ab32503** in HeLa whole cell lysate

Purified **ab32503** immunoprecipitating Bax in HeLa lysates. For western blotting, the primary antibody used was purified **ab32503** at 1/1000 dilution. Ab131366 VeriBlot for IP (HRP) was used for detection at 1/1000 dilution. Capture antibody was used at a 1/20 dilution. Blocking and diluting buffer used was 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32503**).




Western blot - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

**All lanes :** Anti-Bax antibody [E63] (**ab32503**) at 1/1000 dilution

**Lane 1 :** Wild-type HAP1 cell lysate
**Lane 2 :** BAX knockout HAP1 cell lysate

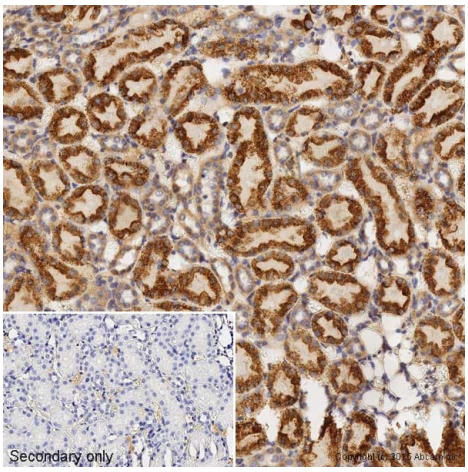
Lysates/proteins at 20 µg per lane.

**Predicted band size:** 21 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab32503**).

**Lanes 1 - 2:** Merged signal (red and green). Green - **ab32503** observed at 20 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

**ab32503** was shown to recognize Bax in wild-type HAP1 cells, along with additional cross-reactive bands. Wild-type and Bax knockout samples were subjected to SDS-PAGE. **ab32503** and **ab8245** (loading control to GAPDH) were diluted at 1/1000 and 1/10 000 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 1/10 000 dilution for 1 h at room temperature before imaging.

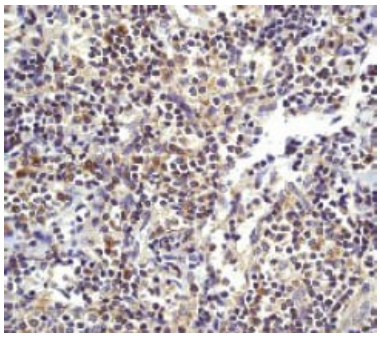



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

IHC image of **ab32503** staining Bax in rat kidney formalin fixed paraffin embedded tissue sections, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with **ab32503**, 1:250 dilution, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. No primary antibody was used in the secondary only control (shown on the inset).
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and

sodium azide (**ab32503**).

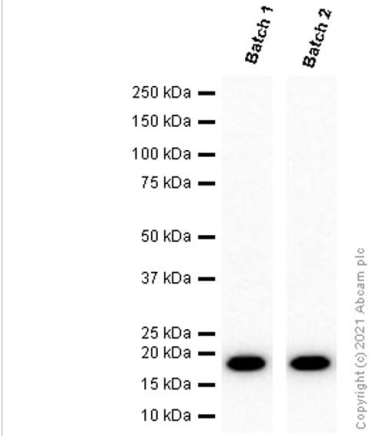


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

Immunohistochemical analysis of paraffin-embedded human lymph node using anti-Bax Rabbit Monoclonal Antibody (**ab32503**) at 1/250 dilution.

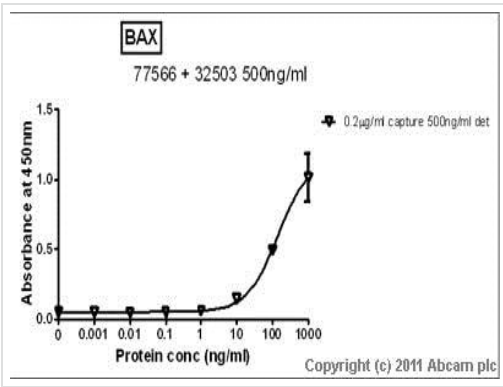
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32503**).

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.




Western blot - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

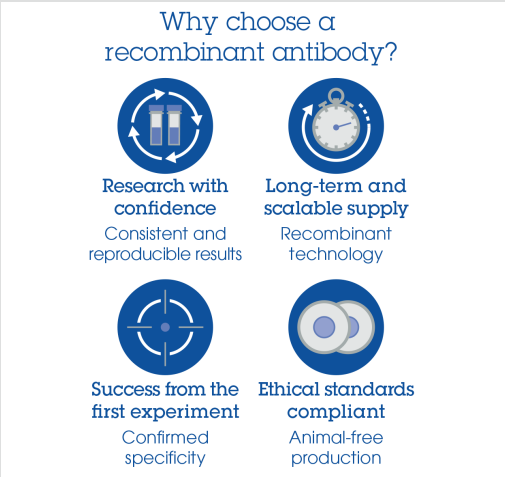
This data was developed using **ab32503**, the same antibody clone in a different buffer formulation. Different batches of **ab32503** were tested on HeLa (Human cervix adenocarcinoma epithelial cell) lysate at 0.1 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 18 kDa.




Sandwich ELISA - Anti-Bax antibody [E63] - BSA and Azide free (ab216985)

Standard Curve for Bax (Analyte: Recombinant human Bax protein (tagged) **ab85157**) dilution range 1pg/ml to 1ug/ml using Capture Antibody **Mouse monoclonal [2D2] to Bax - BSA and Azide free (ab77566)** at 0.2ug/ml and Detector Antibody **Rabbit monoclonal [E63] to Bax (ab32503)** at 0.5ug/ml Concentration of **ab32503** may vary from lot to lot; please use this curve as guideline.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32503**).




Anti-Bax antibody [E63] - BSA and Azide free

(ab216985)

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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