# abcam

### Product datasheet

# Anti-Bcl-XL antibody [E18] ab32370



重组 RabMAb

★★★★★ 10 Abreviews 211 References 18 图像

概述

产品名称 Anti-Bcl-XL抗体[E18]

描述 兔单克隆抗体[E18] to Bcl-XL

宿主 Rabbit

特异性 This antibody should recognize Bcl-XL, Bcl-xS and Bcl-x(beta) as the immunogen sequence is

common to them. The antibody does not cross-react with other Bcl-2 family members.

经测试应用 适用于: WB, IHC-P, ICC/IF, IP, Flow Cyt (Intra)

种属反应性 与反应: Mouse, Rat, Human

预测可用于: Pig 📤

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: Jurkat, K562, C6, RAW264.7, PC-12 NIH/3T3 cell lysates. IHC-P: Human endometrium and

prostate carcinoma tissues. ICC/IF: HepG2, NIH/3T3, C6 and HeLa cells. Flow Cyt (intra): DU145

and Jurkat cells. IP: Jurkat whole cell lysate (ab7899).

常规说明 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.

性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

 $K_D = 6.50 \times 10^{-11} M$ 解离常数(Kn)

10-11



#### Learn more about K<sub>D</sub>

**存储溶液** pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

纯**度** Protein A purified

 克隆
 单克隆

 克隆编号
 E18

 同种型
 IgG

#### 应用

如标

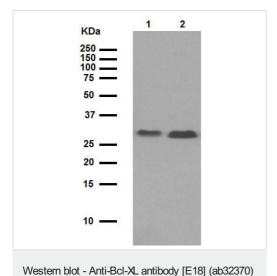
## The Abpromise guarantee Abpromise™承诺保证使用ab32370于以下的经测试应用

"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
WB	**** <u>(8)</u>	1/1000. Detects a band of approximately 26 kDa.
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See <b>IHC antigen retrieval protocols</b> .
ICC/IF	<b>★★★★★ (1)</b>	1/100 - 1/500.
IP		1/10 - 1/30.
Flow Cyt (Intra)		1/20 - 1/100.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

1000	
功能	Potent inhibitor of cell death. Inhibits activation of caspases (By similarity). Appears to regulate cell death by blocking the voltage-dependent anion channnel (VDAC) by binding to it and preventing the release of the caspase activator, CYC1, from the mitochondrial membrane. Isoform Bcl-X(S) promotes apoptosis.
组织 <b>特异性</b>	Bcl-X(S) is expressed at high levels in cells that undergo a high rate of turnover, such as developing lymphocytes. In contrast, $Bcl-X(L)$ is found in tissues containing long-lived postmitotic cells, such as adult brain.
序列相似性	Belongs to the Bcl-2 family.
结 <b>构域</b>	The BH4 motif is required for anti-apoptotic activity. The BH1 and BH2 motifs are required for both heterodimerization with other Bcl-2 family members and for repression of cell death.
翻译后修饰	Proteolytically cleaved by caspases during apoptosis. The cleaved protein, lacking the BH4 motif, has pro-apoptotic activity.
细胞定位	Mitochondrion membrane. Nucleus membrane. Mitochondrial membranes and perinuclear

#### 图片



**All lanes :** Anti-Bcl-XL antibody [E18] (ab32370) at 1/1000 dilution (purified)

Lane 1 : Jurkat cell lysate

Lane 2 : K562 cell lysate

Lysates/proteins at 20 µg per lane.

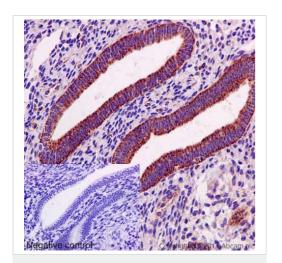
#### Secondary

**All lanes :** Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Observed band size: 26 kDa

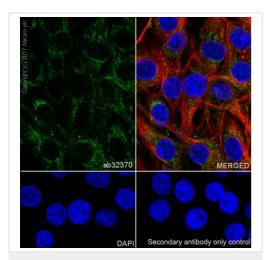
Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



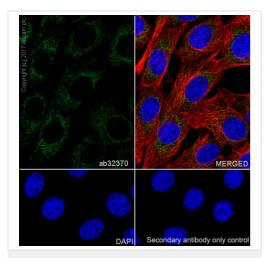
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Bcl-XL antibody [E18] (ab32370)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human endometrium tissue labelling BcI-XL with purified ab32370 at 1/2000. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. <a href="mailto:ab97051">ab97051</a>, a HRP-conjugated goat anti-rabbit HRP (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



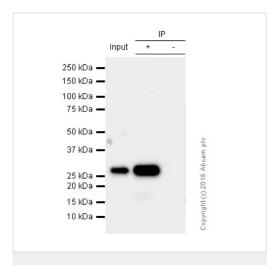
Immunocytochemistry/ Immunofluorescence - Anti-BcI-XL antibody [E18] (ab32370)

Immunocytochemistry/ Immunofluorescence analysis of C6(Rat glial tumor glial cell) cells labeling Bcl-XL with purified ab32370 at 1/100 dilution (1.42 µg/ml). Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 µg/ml). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 dilution (2 µg/ml) dilution. DAPI was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunocytochemistry/ Immunofluorescence - Anti-Bcl-XL antibody [E18] (ab32370)

Immunocytochemistry/ Immunofluorescence analysis of NIH/3T3(Mouse embryonic fibroblast) cells labeling Bcl-XL with purified ab32370 at 1/100 dilution (1.42  $\mu$ g/ml). Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5  $\mu$ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 dilution (2  $\mu$ g/ml) dilution. DAPI was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunoprecipitation - Anti-Bcl-XL antibody [E18] (ab32370)

ab32370 (purified) at 1/20 dilution immunoprecipitating Bcl-XL in NIH/3T3 whole cell lysate.

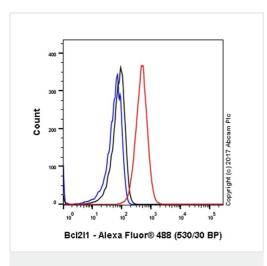
Lane 1 (input): NIH/3T3(Mouse embryonic fibroblast) whole cell lysate  $10\mu g$ 

Lane 2 (+): ab32370 & NIH/3T3 whole cell lysate

Lane 3 (-): Rabbit monoclonal  $\lg G$  (ab172730) instead of ab32370 in NIH/3T3 whole cell lysate

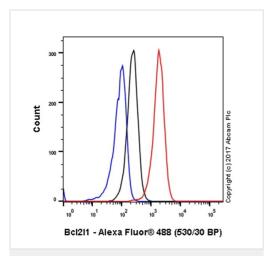
For western blotting, ab32370 at 1/500 dilution (0.28  $\mu$ g/ml) and VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1/1000 dilution.

Blocking and diluting buffer: 5% NFDM /TBST.



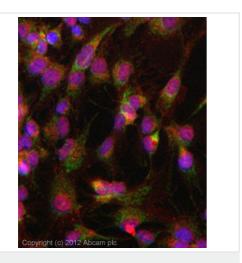
Flow Cytometry (Intracellular) - Anti-Bcl-XL antibody [E18] (ab32370)

Intracellular Flow Cytometry analysis of NIH/3T3 (Mouse embryonic fibroblast) cells labelling Bcl-XL with ab32370 at 1/20 dilution (7.1  $\mu$ g/ml) (Red). Cells were fixed with 4% paraformaldehyde . Goat anti rabbit lgG (Alexa Fluor  $^{\rm @}$  488, <u>ab150077</u>) was used as the secondary antibody at 1/2000 dilution. Isotype control - Rabbit monoclonal lgG (<u>ab172730</u>) (Black). Unlabelled control - Unlabelled cells (Blue).



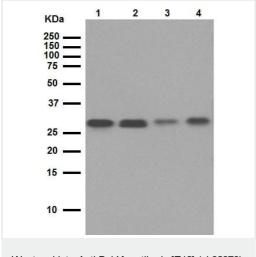
Flow Cytometry (Intracellular) - Anti-Bcl-XL antibody [E18] (ab32370)

Intracellular Flow Cytometry analysis of C6 (Rat glial tumor glial cell) cells labelling Bcl-XL with ab32370 at 1/20 dilution (7.1  $\mu$ g/ml) (Red). Cells were fixed with 4% paraformaldehyde . Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/2000 dilution. Isotype control - Rabbit monoclonal lgG (**ab172730**) (Black). Unlabeled control - Unlabelled cells (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-Bcl-XL antibody [E18] (ab32370)

ICC/IF image of unpurified ab32370 stained HepG2 cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab32370, 1/100) overnight at +4°C. The secondary antibody (green) was  $\underline{ab96899}$ , goat  $\underline{anti-rabbit}\,\underline{DyLight}^{@}\underline{488}$  (IgG; H+L) used at a 1/250 dilution for 1h. Alexa Fluor  $^{@}$  594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Western blot - Anti-Bcl-XL antibody [E18] (ab32370)

**All lanes :** Anti-Bcl-XL antibody [E18] (ab32370) at 1/1000 dilution (purified)

Lane 1: C6 cell lysate

Lane 2: RAW264.7 cell lysate

Lane 3: PC-12 cell lysate

Lane 4: NIH/3T3 cell lysate

Lysates/proteins at 20 µg per lane.

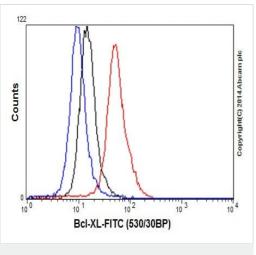
#### Secondary

**All lanes :** Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Observed band size: 26 kDa

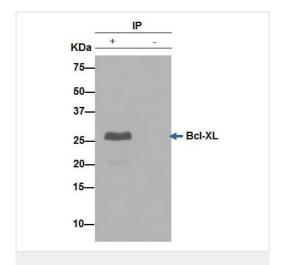
Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Flow Cytometry (Intracellular) - Anti-Bcl-XL antibody [E18] (ab32370)

Intracellular Flow Cytometry analysis of Jurkat cells labelling Bcl-XL with purified ab32370 at 1/20 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



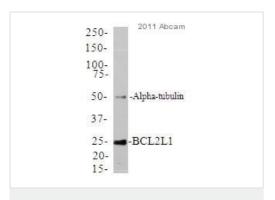
Immunoprecipitation - Anti-Bcl-XL antibody [E18]

(ab32370)

ab32370 (purified) at 1/30 immunoprecipitating Bcl-XL in Jurkat cell lysate (Lane 1). Lane 2 - PBS. For western blotting, a HRP-conjugated anti-rabbit lgG, specific to the non-reduced form of lgG was used as the secondary antibody (1/1500).

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-Bcl-XL antibody [E18] (ab32370) Image courtesy of an anonymous Abreview.

Anti-Bcl-XL antibody [E18] (ab32370) at 1/500 dilution (unpurified) + whole cell lysate prepared from a clinical sample of human breast cancer cells at 20  $\mu g$ 

#### Secondary

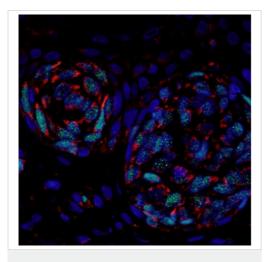
Goat anti-rabbit IgG-HRP at 1/1000 dilution

Developed using the ECL technique.

Observed band size: 26 kDa

Exposure time: 15 minutes

Patient recieved anthracycline and taxane neoadjuvant chemotherapy.

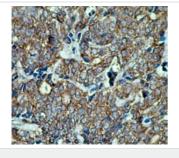


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Bcl-XL antibody [E18] (ab32370)

Image from Medic S & Ziman M, PLoS One. 2010 Apr 22;5(4):e9977. Fig 5.; doi:10.1371/journal.pone.0009977; April 22, 2010, PLoS ONE 5(4): e9977.

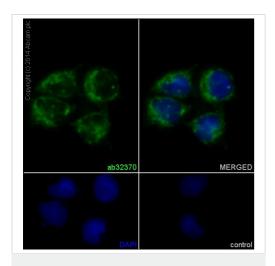
Immunohistochemistry of human primary melanoma, staining Bcl-XL (red) with unpurified ab32370.

Antigen retrieval was performed in EDTA/Tris buffer (pH 8) before being blocked with 10%NGS for one hour at room temperature. Samples were incubated with primary antibody (1/50) at room temperature for one hour. An AlexaFluor®-conjugated anti-rabbit lgG was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Bcl-XL antibody [E18] (ab32370)

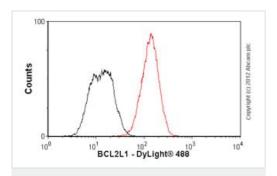
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human prostate carcinoma tissue labelling Bcl-XL with unpurified ab32370 at 1/50.



Immunocytochemistry/ Immunofluorescence - Anti-Bcl-XL antibody [E18] (ab32370)

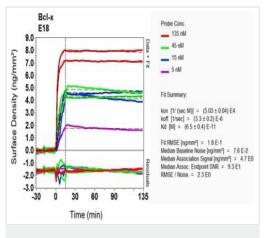
Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling BcI-XL with purified ab32370 at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. <a href="mailto:ab150077">ab150077</a>, a goat anti-rabbit Alexa Fluor<sup>®</sup> 488 (lgG; 1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

Control: primary antibody (1/500) and secondary antibody, **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).



Flow Cytometry (Intracellular) - Anti-Bcl-XL antibody [E18] (ab32370)

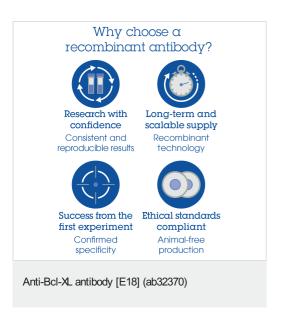
Overlay histogram showing DU145 cells stained with unpurified ab32370 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32370, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was a goat anti-rabbit DyLight® 488 (lgG; H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (1µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed.



Ol-RD Scanning - Anti-Bcl-XL antibody [E18] (ab32370)

Equilibrium disassociation constant ( $K_D$ ) Learn more about  $K_D$ 

#### Click here to learn more about KD



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