abcam

Product datasheet

Anti-Bim antibody [Y36] ab32158



★★★★ 6 Abreviews 93 References

画像数 12

製品の概要

製品名 Anti-Bim antibody [Y36]

製品の詳細 Rabbit monoclonal [Y36] to Bim

由来種 Rabbit

特異性 Based on the sequence homology of the immunogen, this antibody is likely to detect all Bim

アプリケーション 適用あり: Flow Cyt (Intra), WB, IHC-P, IP, ICC/IF

種交差性 交差種: Mouse, Rat, Human

免疫原 Synthetic peptide within Human Bim aa 1-100. The exact sequence is proprietary.

(Peptide available as ab179844)

ポジティブ・コントロール WB: Raji, Jurkat, A431, Molt-4, A20, MEF, Raw264.7 and PC-12 cell lysate; Human and mouse

> thymus, mouse and rat spleen tissue lysate. IHC: breast carcinoma tissue. ICC/IF: A20 and Raji cells. Flow Cyt (intra): A431 and Raji whole cell lysate. HAP1-WT cells. IP: Raji whole cell lysate.

特記事項 Mouse and Rat species are recommended based on WB results, we do not guarantee

IHC-P for Mouse and Rat.

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**.

製品の特性

製品の状態

保存方法 Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 49% PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA

精製度 Protein A purified

ポリ/モノ モノクローナル

ウローン名 Y36 アイソタイプ IgG

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab32158の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/50 - 1/100.
WB	★★★★ ★ (5)	1/500 - 1/2000. Detects a band of approximately 22 kDa (predicted molecular weight: 22 kDa). Can be blocked with Bim peptide (ab179844).
IHC-P		1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. Mouse and Rat species are recommended based on WB results, we do not guarantee IHC-P for Mouse and Rat.
IP	****(1)	1/40 - 1/50.
ICC/IF		1/100 - 1/250.

ターゲット情報

機能 Induces apoptosis. Isoform BimL is more potent than isoform BimEL. Isoform Bim-alpha1,

isoform Bim-alpha2 and isoform Bim-alpha3 induce apoptosis, although less potent than the

isoforms BimEL, BimL and BimS. Isoform Bim-gamma induces apoptosis.

組織特異性 Isoform BimL and isoform BimS are the predominant isoforms and are

ubiquitously expressed with a tissue-specific variation. Isoform Bim-gamma is most abundantly expressed in small intestine and colon, and in lower levels in spleen, prostate, testis, heart, liver

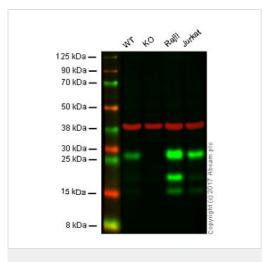
and kidney.

配列類似性 Belongs to the Bcl-2 family.

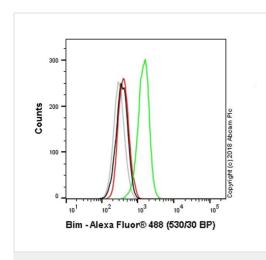
ドメイン The BH3 motif is required for Bcl-2 binding and cytotoxicity.

細胞内局在 Mitochondrion and Endomembrane system. Associated with intracytoplasmic membranes.

画像



Western blot - Anti-Birn antibody [Y36] (ab32158)



Flow Cytometry (Intracellular) - Anti-Bim antibody [Y36] (ab32158)

Lane 1: Wild type HAP1 whole cell lysate (20 µg)

Lane 2: Bim knockout HAP1 whole cell lysate (20 µg)

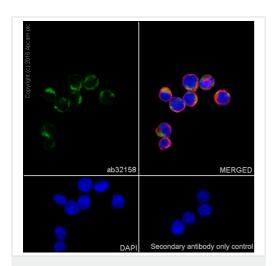
Lane 3: Raji whole cell lysate (20 µg)

Lane 4: Jurkat whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab32158 observed at 25 kDa. Red - loading control, **ab9484**, observed at 37 kDa.

ab32158 was shown to specifically react with Bim when Bim knockout samples were used. Wild-type and Bim knockout samples were subjected to SDS-PAGE. ab32158 and <u>ab9484</u> (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/500 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye[®] 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse IgG H&L (IRDye[®] 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.

Overlay histogram showing HAP1 wildtype (green line) and HAP1-BCL2L11 knockout cells (red line) stained with ab32158. The cells were fixed 80% methanol (5 min), and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab32158, 1µg/ml) for 30 min at 22°C. The secondary antibody used was Alexa Fluor[®] 488 goat anti-rabbit lgG (H&L) presorbed (ab150081) at 1/2000 dilution for 30 min at 22°C. A rabbit lgG1 isotype control antibody (ab172730) was used at the same concentration and conditions as the primary antibody (HAP1 wildtype - black line, HAP1-BCL2L11 knockout - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity). Acquisition of >5,000 events were collected using a 50 mW Blue laser (488nm) and 530/30 bandpass filter.

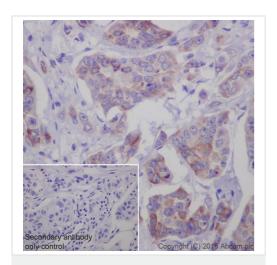


Immunocytochemistry/ Immunofluorescence - Anti-Bim antibody [Y36] (ab32158)

Immunocytochemistry/Immunofluorescence analysis of Raji (Human Burkitt's lymphoma cell line) labeling Bim with ab32158 at a dilution of 1/250. Cells were fixed with 100% methanol. Ab150077 (1/1000) was used as the secondary antibody. Cells were co-stained with ab7291, a mouse anti-tubulin antibody (1/200) using ab150120 as the secondary. Nuclei were counterstained with DAPI (blue).

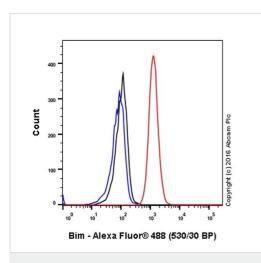
Secondary antibody only control, cells without incubation with the primary antibody was used as negative control.

Confocal image showing cytoplasmic staining on Raji cell line



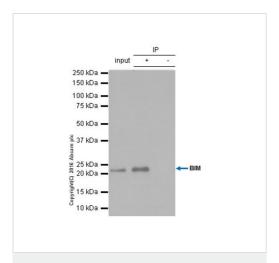
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Bim antibody [Y36] (ab32158)

ab32158 staining Bim in human breast cancer tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffinembedded sections). Antigen retrieval was by heat mediated antigen retrieval using Tris/EDTA Buffer, PH9 (ab93684). Samples were incubated with primary antibody (1/100 in blocking buffer) and a Biotin-conjugated Donkey anti-rabbit IgG polyclonal (1/500) was used as the secondary antibody. Cytoplasmic staining can be seen in the human breast cancer cells. Hematoxylin was used as a counter stain.



Flow Cytometry (Intracellular) - Anti-Bim antibody [Y36] (ab32158)

Intracellular Flow Cytometry analysis of Raji (human Burkitt's lymphoma) whole cell lysate labeling Bim with ab32158 at 1/100 (red). Cells were fixed with 4% paraformaldehyde. An Alexa Fluorr® 488)-conjugated goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG (ab172730). Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Immunoprecipitation - Anti-Bim antibody [Y36] (ab32158)

Ab32158 at 1/50 immunoprecipitating Bim in Raji (human Burkitt's lymphoma) whole cell lysate.

Lane 1 (input): Raji whole cell lysate (10µg)

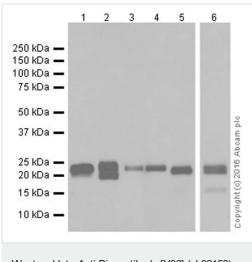
Lane 2 (+): ab32158 + Raji whole cell lysate.

Lane 3 (-): Rabbit monoclonal lgG (<u>ab172730</u>) instead of ab32158 in Raji whole cell lysate.

For western blotting, ab32158 (1/1000) was used as the primary antibody and <u>ab131366</u> VeriBlot for IP Detection Reagent (HRP) was used for detection (1/10 000).

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-Birn antibody [Y36] (ab32158)

All lanes: Anti-Bim antibody [Y36] (ab32158) at 1/2000 dilution

Lane 1: Raji (human Burkitt's lymphoma) whole cell lysate

Lane 2: A431 (human epidermoid carcinoma) whole cell lysate

Lane 3 : Molt-4 (human acute lymphoblastic leukemia) whole cell

lysate

Lane 4: Human thymus tissue lysate

Lane 5: Mouse thymus tissue lysate

Lane 6: A20 (mouse reticulum cell sarcoma) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 22 kDa

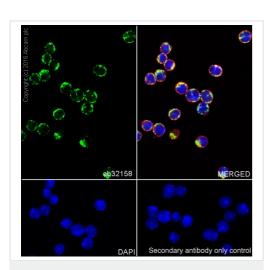
Observed band size: 22, 18 kDa

Exposure time: Lane 1-5:3 minutes; Lane 6:2 seconds

Blocking/Diluting buffer and concentration: 5% NFDM /TBST

The observed molecular weight is consistent with the literature

(PMID: 24872388)

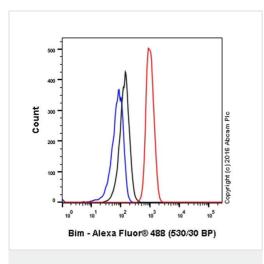


Immunocytochemistry/ Immunofluorescence - Anti-Bim antibody [Y36] (ab32158)

Immunocytochemistry/Immunofluorescence analysis of A20 (Mouse reticulum sarcoma cell line) labeling Bim with ab32158 at a dilution of 1/250. Cells were fixed with 100% methanol. Ab150077 (1/1000) was used as the secondary antibody. Cells were co-stained with ab7291, a mouse anti-tubulin antibody (1/200) using ab150120 as the secondary. Nuclei were counterstained with DAPI (blue).

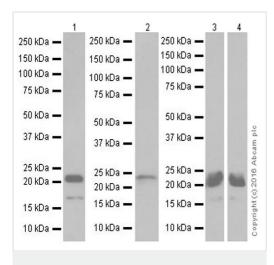
Secondary antibody only control, cells without incubation with the primary antibody was used as negative control.

Confocal image showing cytoplasmic staining on A20 cell line



Flow Cytometry (Intracellular) - Anti-Bim antibody [Y36] (ab32158)

Intracellular Flow Cytometry analysis of A431 (human epidermoid carcinoma) cells labelling Bim with ab32158 at 1/50 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. An Alexa Fluorr®488-conjugated goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Western blot - Anti-Bim antibody [Y36] (ab32158)

All lanes: Anti-Bim antibody [Y36] (ab32158) at 1/2000 dilution

Lane 1: Mouse spleen tissue lysate

Lane 2: Rat spleen tissue lysate

Lane 3: PC-12 (rat adrenal gland pheochromocytoma) whole cell

lysate

Lane 4: Raw264.7 (mouse abelson murine leukemia virus-induced

tumor) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at

1/100000 dilution

Predicted band size: 22 kDa

Observed band size: 22, 18 kDa

Exposure time: Lane 1-3: 3 minutes; Lane 4: 10 seconds

Blocking/Diluting buffer and concentration: 5% NFDM /TBST

The observed molecular weight is consistent with the literature

(PMID: 24872388)

Anti-Bim antibody [Y36] (ab32158) at 1/500 dilution + Whole cell lysates prepared from human jurkat cells at 200000 cells

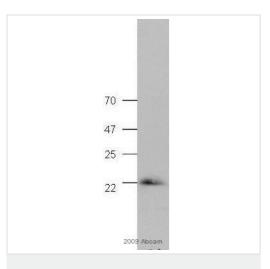
Secondary

HRP conjugated Donkey polyclonal to rabbit IgG at 1/2000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

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

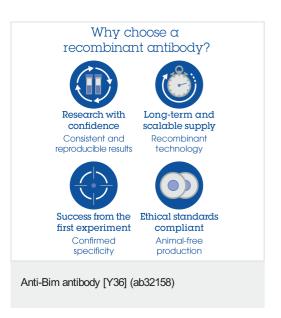


Western blot - Anti-Bim antibody [Y36] (ab32158)

This image is courtesy of an anonymous abreview.

Exposure time: 30 seconds

Primary diluted in PBS (5% BSA + 0.1% tween20) and incubated with sample for 1 hour and 30 minutes at 20°C.



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