abcam

Product datasheet

Anti-Bcl-2 antibody [EPR17509] ab182858

KO 評価済 リコンビナント RabMAb

13 Abreviews 458 References 画像数 11

製品の概要

製品名	Anti-Bcl-2 antibody [EPR17509]
製品の詳細	Rabbit monoclonal [EPR17509] to Bcl-2
由来種	Rabbit
アプリケーション	**適用あり:** Flow Cyt (Intra), WB, IHC-P **適用なし:** ICC/IF
種交差性	**交差種:** Mouse, Human
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	WB: Human tonsil and thymus lysates; Jurkat, U-937, THP-1, HeLa, C2C12, WEHI -3 and NIH/3T3 whole cell lysates; Mouse brain, heart, kidney and spleen lysates; Human fetal kidney and fetal spleen lysates; Wild-type Hap1 cell lysate. IHC-P: Human tonsil tissue, Human endometrial cancer tissue, Mouse spleen tissue. Flow Cyt (intra): Jurkat cells.
特記事項	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	Liquid
保存方法	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.
バッファー	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
精製度	Protein A purified
ポリ/モノ	モノクローナル

クローン名	EPR17509
アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、次のテスト済みアプリケーションにおけるab182858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)		1/250.
WB	★★★★★ (5)	1/2000. Detects a band of approximately 26 kDa (predicted molecular weight: 26 kDa).
IHC-P	★★★★☆ (7)	1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

追加情報 Is unsuitable for ICC/IF.

ターゲット情報

機能 Suppresses apoptosis in a variety of cell systems including factor-dependent lymphohematopoietic and neural cells. Regulates cell death by controlling the mitochondrial membrane permeability. Appears to function in a feedback loop system with caspases. Inhibits caspase activity either by preventing the release of cytochrome c from the mitochondria and/or by binding to the apoptosis-activating factor (APAF-1). May attenuate inflammation by impairing NLRP1-inflammasome activation, hence CASP1 activation and IL1B release (PubMed:17418785).

組織特異性 Expressed in a variety of tissues.

関連疾患 A chromosomal aberration involving BCL2 has been found in chronic lymphatic leukemia. Translocation t(14;18)(q32;q21) with immunoglobulin gene regions. BCL2 mutations found in non-Hodgkin lymphomas carrying the chromosomal translocation could be attributed to the Ig somatic hypermutation mechanism resulting in nucleotide transitions.

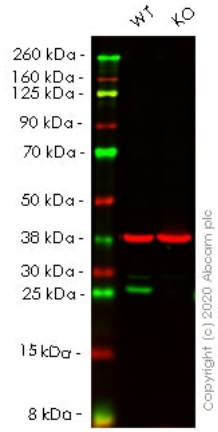
配列類似性 Belongs to the Bcl-2 family.

ドメイン BH1 and BH2 domains are required for the interaction with BAX and for anti-apoptotic activity. The BH4 motif is required for anti-apoptotic activity and for interaction with RAF1 and EGLN3. The loop between motifs BH4 and BH3 is required for the interaction with NLRP1.

翻訳後修飾 Phosphorylation/dephosphorylation on Ser-70 regulates anti-apoptotic activity. Growth factor-stimulated phosphorylation on Ser-70 by PKC is required for the anti-apoptosis activity and occurs during the G2/M phase of the cell cycle. In the absence of growth factors, BCL2 appears to be phosphorylated by other protein kinases such as ERKs and stress-activated kinases. Phosphorylated by MAPK8/JNK1 at Thr-69, Ser-70 and Ser-87, wich stimulates starvation-induced autophagy. Dephosphorylated by protein phosphatase 2A (PP2A).
Proteolytically cleaved by caspases during apoptosis. The cleaved protein, lacking the BH4 motif, has pro-apoptotic activity, causes the release of cytochrome c into the cytosol promoting further caspase activity.
Monoubiquitinated by PARK2, leading to increase its stability. Ubiquitinated by SCF(FBXO10),

	leading to its degradation by the proteasome.
細胞内局在	Mitochondrion outer membrane. Nucleus membrane. Endoplasmic reticulum membrane.

画像



Western blot - Anti-Bcl-2 antibody [EPR17509] (ab182858)

All lanes : Anti-Bcl-2 antibody [EPR17509] (ab182858) at 1/2000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : BCL2 knockout HeLa cell lysate

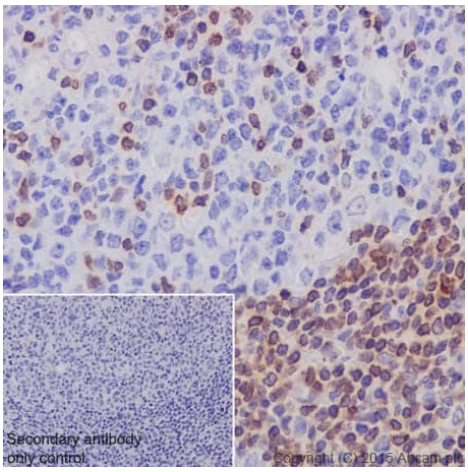
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 26 kDa
Observed band size: 26 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab182858 observed at 26 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab182858 was shown to react with Bcl-2 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab255364** (knockout cell lysate **ab263752**) was used. Wild-type HeLa and BCL2 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. ab182858 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 2000 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-Bcl-2 antibody [EPR17509] (ab182858)

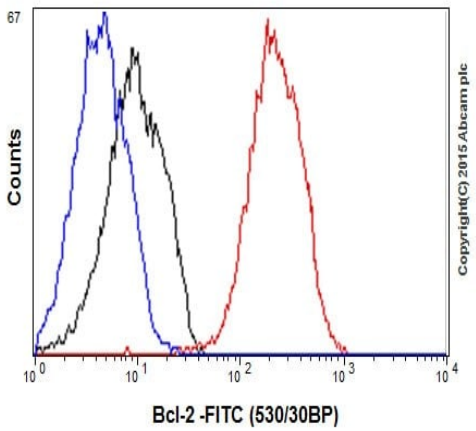
Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling Bcl-2 with ab182858 at 1/1000 followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500.

Cytoplasm, nuclear membrane and nucleus staining on lymphocytes of Human tonsil tissue is observed.

Counter stained with Hematoxylin.

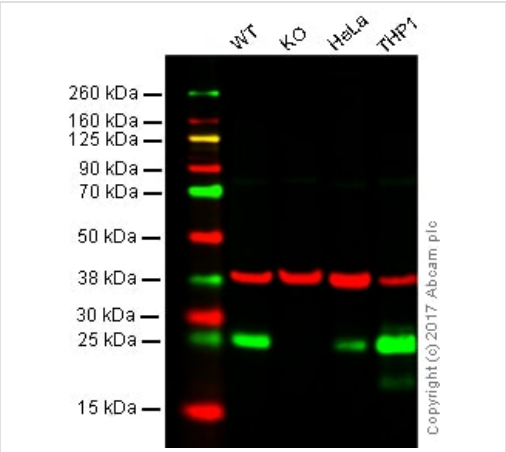
Negative control: Used PBS instead of primary antibody followed by **ab97051** at 1/500.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-Bcl-2 antibody [EPR17509] (ab182858)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling Bcl-2 with ab182858 at 1/250 (red) compared with a rabbit monoclonal IgG isotype control (**ab172730**) (black) and a unlabelled control (cells without incubation with primary antibody and secondary antibody (blue)). Goat anti rabbit IgG (FITC) at 1/500 was used as the secondary antibody.



Western blot - Anti-Bcl-2 antibody [EPR17509] (ab182858)

All lanes : Anti-Bcl-2 antibody [EPR17509] (ab182858) at 1 µg/ml

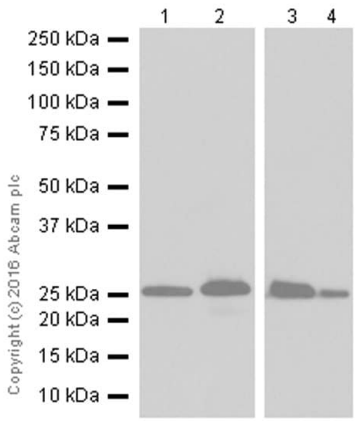
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : BCL2 knockout HAP1 whole cell lysate
Lane 3 : HeLa whole cell lysate
Lane 4 : THP-1 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 26 kDa
Observed band size: 26 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab182858 observed at 26 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab182858 was shown to specifically react with BCL2 when BCL2 knockout samples were used. Wild-type and BCL2 knockout samples were subjected to SDS-PAGE. Ab182858 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1 ug/ml 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-Bcl-2 antibody [EPR17509] (ab182858)

All lanes : Anti-Bcl-2 antibody [EPR17509] (ab182858) at 1/10000 dilution

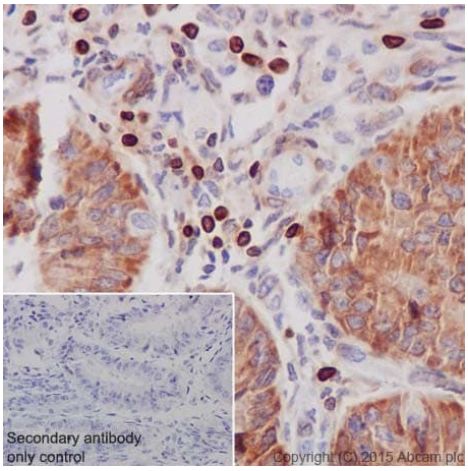
Lane 1 : NIH/3T3 (mouse embryo fibroblast cell line) whole cell lysate at 20 µg
Lane 2 : WEHI-3 (mouse leukemia cell line) whole cell lysate at 20 µg
Lane 3 : Mouse hippocampus at 10 µg
Lane 4 : Mouse heart at 10 µg

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) at 1/2000 dilution

Predicted band size: 26 kDa
Observed band size: 26 kDa

Exposure time: 8 seconds

Blocking/Diluting buffer 5% NFDM/TBST



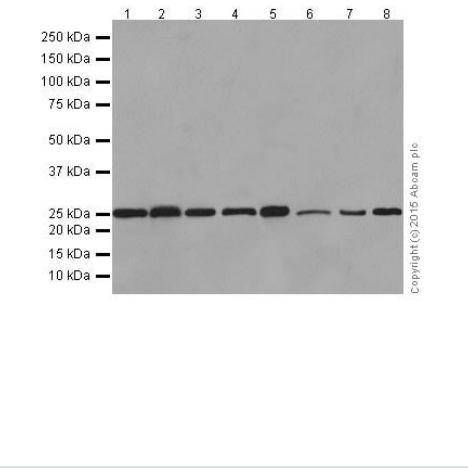
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [EPR17509] (ab182858)

Immunohistochemical analysis of paraffin-embedded Human endometrial cancer tissue labeling Bcl-2 with ab182858 at 1/1000 followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500.

Cytoplasm, nuclear membrane and nucleus staining on lymphocytes and cancer cells of Human endometrial cancer tissue is observed.

Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary antibody followed by **ab97051** at 1/500.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Western blot - Anti-Bcl-2 antibody [EPR17509] (ab182858)

All lanes : Anti-Bcl-2 antibody [EPR17509] (ab182858) at 1/20000 dilution

Lane 1 : Human tonsil lysate
Lane 2 : Human thymus lysate
Lane 3 : Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysate
Lane 4 : U-937 (Human histiocytic lymphoma cells) whole cell lysate
Lane 5 : THP-1 (Human monocytic leukemia cells) whole cell lysate
Lane 6 : HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate
Lane 7 : C2C12 (Mouse myoblast cell line) whole cell lysate
Lane 8 : WEHI-3 (Mouse leukemia cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

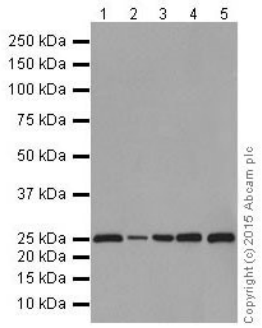
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/1000 dilution

Developed using the ECL technique.

Predicted band size: 26 kDa
Observed band size: 26 kDa

Exposure time: 1 minute

Blocking and diluting buffer was 5% NFDM /TBST.



Western blot - Anti-Bcl-2 antibody [EPR17509] (ab182858)

All lanes : Anti-Bcl-2 antibody [EPR17509] (ab182858) at 1/2000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse heart lysate
Lane 3 : Mouse kidney lysate
Lane 4 : Mouse spleen lysate
Lane 5 : NIH/3T3 (Mouse embryo fibroblast cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

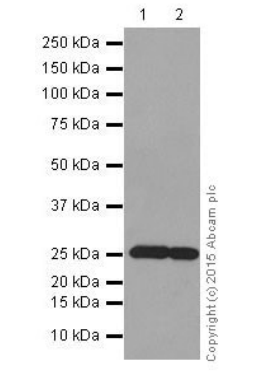
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/1000 dilution

Developed using the ECL technique.

Predicted band size: 26 kDa
Observed band size: 26 kDa

Exposure time: 3 minutes

Blocking and diluting buffer was 5% NFDM /TBST.



Western blot - Anti-Bcl-2 antibody [EPR17509] (ab182858)

All lanes : Anti-Bcl-2 antibody [EPR17509] (ab182858) at 1/2000 dilution

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

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

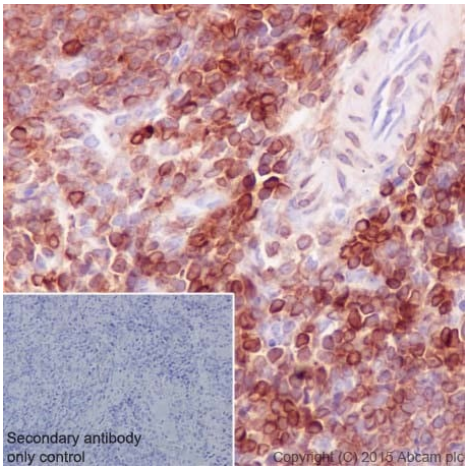
Developed using the ECL technique.

Predicted band size: 26 kDa

Observed band size: 26 kDa

Exposure time: 3 minutes

Blocking and diluting buffer was 5% NFDM /TBST.



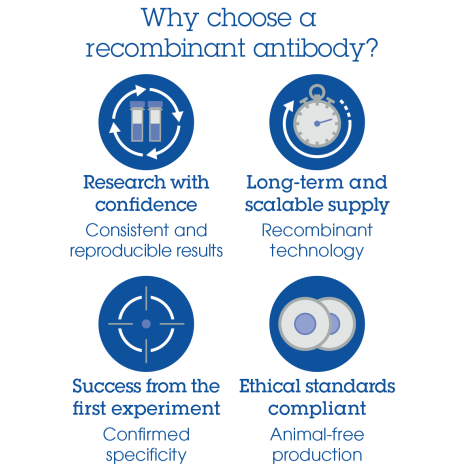
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [EPR17509] (ab182858)

Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labeling Bcl-2 with ab182858 at 1/1000 followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500.

Cytoplasm, nuclear membrane and nucleus staining on lymphocytes of Mouse spleen tissue is observed.

Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary antibody followed by **ab97051** at 1/500.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Anti-Bcl-2 antibody [EPR17509] (ab182858)

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