

Anti-Bcl-2 antibody [E17] ab32124

敲除验证
重组
RabMAb

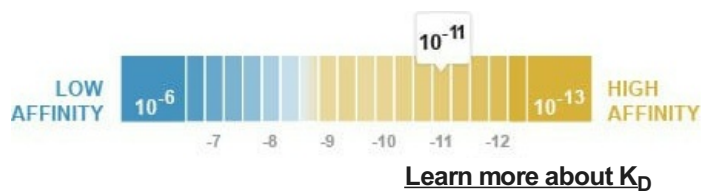
★★★★☆
[14 Abreviews](#)
[1122 References](#)
[16 图像](#)

概述

产品名称	Anti-Bcl-2抗体[E17]
描述	兔单克隆抗体[E17] to Bcl-2
宿主	Rabbit
特异性	<p>This antibody recognises Bcl-2. It does not cross-react with other Bcl-2 family members.</p> <p>Bcl-2 has two isoforms, one is around 26kDa and the other is around 20kDa (PMID: 26009263, PMID: 10400666, PMID: 32377726).</p>
经测试应用	<p>适用于: WB, IHC-P, IP</p> <p>不适用于: Flow Cyt or ICC/IF</p>
种属反应性	与反应: Human
免疫原	<p>Synthetic peptide within Human Bcl-2 aa 50-150. The exact sequence is proprietary.</p> <p>Database link: P10415</p>
阳性对照	<p>WB: MCF7, A431, Jurkat, HeLa, THP-1 and SH-SY5Y cell lysates; Wild type HAP1 whole cell lysate. IHC-P: Human B-cell lymphoma and breast carcinoma tissues; Human UM xenografts; Human salivary glands; Human DLBCL U2932 cell line xenograft tissue. IP: Jurkat whole cell lysate (ab7899).</p>
常规说明	<p>Abcam recommended secondaries - Goat Anti-Rabbit HRP (ab205718) and Goat Anti-Rabbit Alexa Fluor® 488 (ab150077). Or search our wide range of secondary antibodies for use with your experiment.</p> <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.</p> <p>Rat: We have preliminary internal testing data to indicate this antibody may not react with this species. Please contact us for more information.</p>

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.
解离常数 (K_D)	$K_D = 3.00 \times 10^{-11}$ M



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	E17
同种型	IgG

应用

The Abpromise guarantee **Abpromise™**承诺保证使用ab32124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	★★★★★ (7)	1/1000. Detects a band of approximately 26 kDa (predicted molecular weight: 26 kDa). We recommend using ab182858 for murine samples.
IHC-P	★★★★★ (1)	1/250 - 1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols . We do not recommend rat and mouse samples with IHC.
IP	★★★★★ (1)	1/50.

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

功能	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
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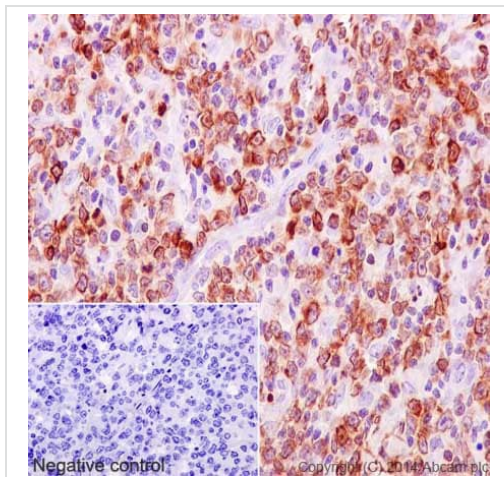
	NLRP1-inflammasome activation, hence CASP1 activation and IL1B release (PubMed:17418785).
组织特异性	Expressed in a variety of tissues.
疾病相关	A chromosomal aberration involving BCL2 has been found in chronic lymphatic leukemia. Translocation t(14;18)(q32;q21) with immunoglobulin gene regions. BCL2 mutations found in non-Hodgkin lymphomas carrying the chromosomal translocation could be attributed to the Ig somatic hypermutation mechanism resulting in nucleotide transitions.
序列相似性	Belongs to the Bcl-2 family.
结构域	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.
翻译后修饰	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.
细胞定位	Mitochondrion outer membrane. Nucleus membrane. Endoplasmic reticulum membrane.

图片

Tissue Microarray (TMA) data for ab32124					
Normal tissue samples			Malignant tissue samples		
Human cardiac muscle	x	Human placenta	✓	Clear cell carcinoma of human kidney	✓
Human cerebrum	x	Human skeletal muscle	x	Human bladder cancer	x
Human colon	x (immune cells ✓)	Human skin	x (immune cells ✓)	Human breast carcinoma	x (immune cells ✓)
Human endometrium	✓	Human spleen	✓	Human cervical carcinoma	x
Human kidney	✓	Human stomach	x (immune cells ✓)	Human colon carcinoma	✓
Human liver	x (immune cells ✓)	Human testis	x	Human endometrial carcinoma	x (immune cells ✓)
Human lung	x (immune cells ✓)	Human thyroid	✓	Human gastric adenocarcinoma	x (immune cells ✓)
Human mammary gland	✓	Human tonsil	✓	Human non-Hodgkin's lymphoma	✓
Human pancreas	x (immune cells ✓)			Human Hodgkin's lymphoma	✓
				Human glioma	✓
				Human hepatocellular carcinoma	x (immune cells ✓)
				Human lung carcinoma	x (immune cells ✓)
				Human ovarian carcinoma	✓
				Human pancreatic carcinoma	x (immune cells ✓)
				Human prostatic hyperplasia	✓
				Human thyroid carcinoma	✓
				Human thymoma	✓

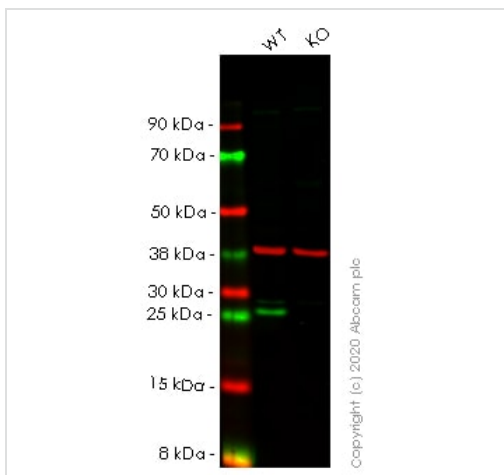
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [E17] (ab32124)

Tissue Microarrays for Anti-Bcl2 antibody [E17] using ab32124 in immunohistochemical analysis. This table provides a detailed overview of positive (tick mark) and negative (cross mark) staining per sample type tested. The sections were pretreated with heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins. The section was incubated with ab32124 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [E17] (ab32124)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human B-cell lymphoma tissue labelling Bcl-2 with purified ab32124 at 1/250. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. [ab97051](#), a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Western blot - Anti-Bcl-2 antibody [E17] (ab32124)

All lanes : Anti-Bcl-2 antibody [E17] (ab32124) at 1/1000 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.

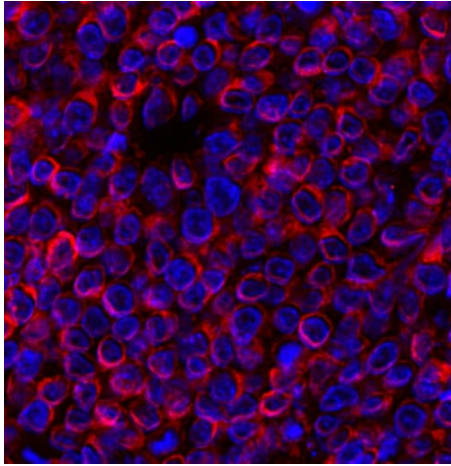
Predicted band size: 26 kDa

Observed band size: 26 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab32124 observed at 26 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) observed at 37 kDa.

ab32124 was shown to react with BCL2 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. ab32124 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.

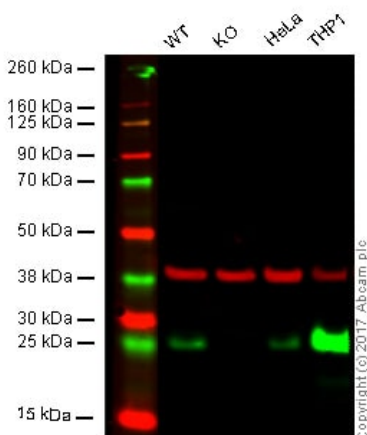
Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [E17] (ab32124)

This image is courtesy of an anonymous Abreview.

Formaldehyde-fixed, paraffin-embedded human DLBCL U2932 cell line xenograft tissue stained for Bcl-2 using ab32124 at 1/200 dilution in immunohistochemical analysis, followed by Goat anti-Rabbit IgG Alexa Fluor® 555.



Western blot - Anti-Bcl-2 antibody [E17] (ab32124)

All lanes : Anti-Bcl-2 antibody [E17] (ab32124)

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

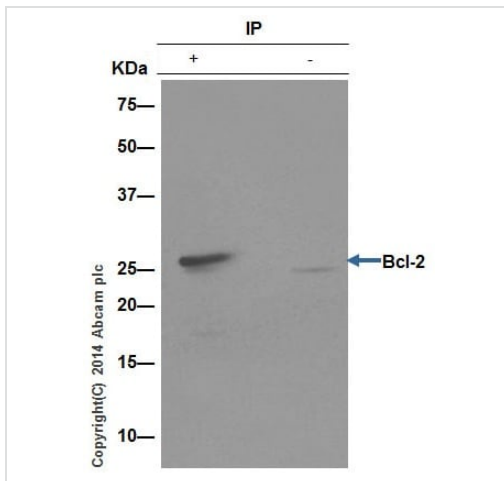
Lysates/proteins at 20 µg per lane.

Predicted band size: 26 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab32124 observed at 26 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab32124 was shown to specifically react with BCL2 when BCL2 knockout samples were used. Wild-type and BCL2 knockout samples were subjected to SDS-PAGE. Ab32124 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 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. 3% milk used as blocking agent.

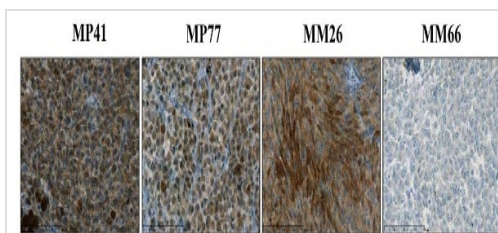


Immunoprecipitation - Anti-Bcl-2 antibody [E17]
(ab32124)

ab32124 (purified) at 1/30 immunoprecipitating Bcl-2 in Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate (Lane 1). Lane 2 - PBS. For western blotting, a HRP-conjugated anti-rabbit IgG, specific to the non-reduced form of IgG was used as the secondary antibody (1/1500).

Blocking buffer and concentration: 5% NFDM/TBST.

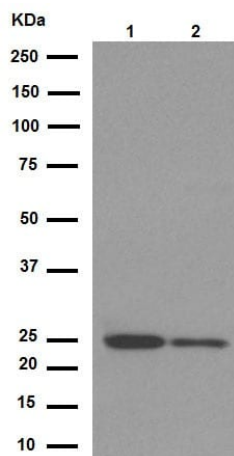
Diluting buffer and concentration: 5% NFDM /TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [E17]
(ab32124)

Image from Némati F et al., PLoS One. 2014;9(1):e80836. Fig 2.; doi: 10.1371/journal.pone.0080836. Reproduced under the Creative Commons license <http://creativecommons.org/licenses/by/4.0/>

Bcl-2 expression determined by immunohistochemical analyses of the 4 human UM xenografts (between 3 to 5 tumors have been studied per condition).



Western blot - Anti-Bcl-2 antibody [E17] (ab32124)

All lanes : Anti-Bcl-2 antibody [E17] (ab32124) at 1/1000 dilution (purified)

Lane 1 : MCF7 (human breast adenocarcinoma cell line) cell lysate

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

Lysates/proteins at 20 µg per lane.

Secondary

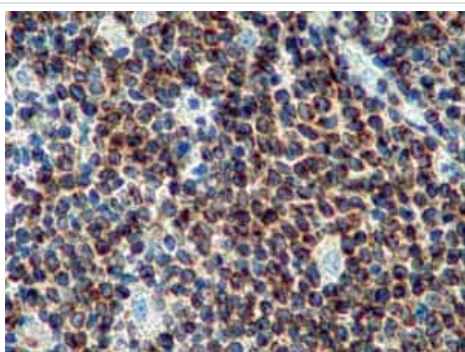
All lanes : Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 26 kDa

Observed band size: 26 kDa

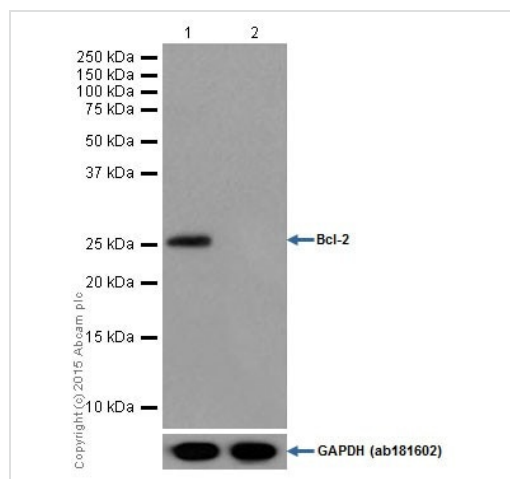
Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [E17] (ab32124)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human B cell lymphoma tissue labelling Bcl-2 with unpurified ab32124. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.



Western blot - Anti-Bcl-2 antibody [E17] (ab32124)

All lanes : Anti-Bcl-2 antibody [E17] (ab32124) at 1/200 dilution

Lane 1 : MCF7 (human breast adenocarcinoma cell line) cell lysate

Lane 2 : SK-BR-3 (human mammary gland adenocarcinoma cell line) cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/50000 dilution

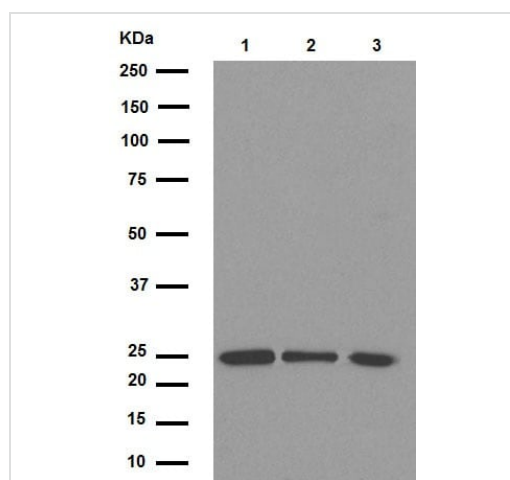
Predicted band size: 26 kDa

Observed band size: 26 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

MCF-7 cells express Bcl-2, while SK-BR-3 cells do not express Bcl-2 (PMID: 18430249)



Western blot - Anti-Bcl-2 antibody [E17] (ab32124)

All lanes : Anti-Bcl-2 antibody [E17] (ab32124) at 1/10000 dilution (purified)

Lane 1 : Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate

Lane 2 : HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate

Lane 3 : SH-SY5Y (human neuroblastoma cell line from bone marrow) cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

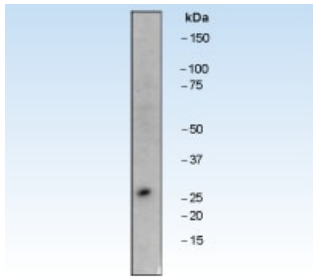
All lanes : Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 26 kDa

Observed band size: 26 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

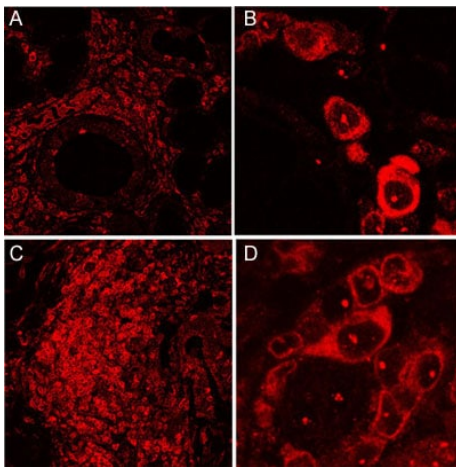


Western blot - Anti-Bcl-2 antibody [E17] (ab32124)

Anti-Bcl-2 antibody [E17] (ab32124) at 1/1000 dilution (unpurified)
+ Jurkat (human T cell leukemia cell line from peripheral blood) cell
lysate

Predicted band size: 26 kDa

Observed band size: 26 kDa



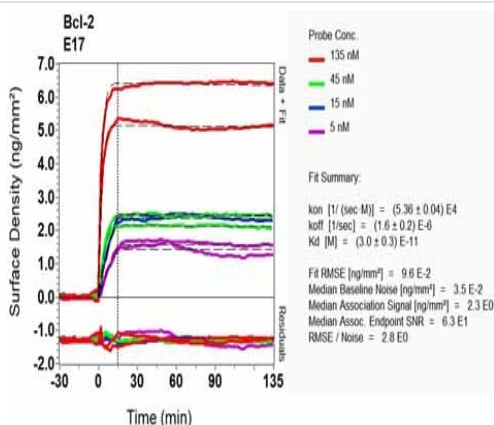
Immunohistochemistry (Formalin/PFA-fixed paraffin-
embedded sections) - Anti-Bcl-2 antibody [E17]
(ab32124)

Image from Szyzko EA et al., Arthritis Res Ther. 2011
Jan 7;13(1):R2. Fig 5.; doi:10.1186/ar3220; 7 January
2011, Arthritis Research & Therapy 2011, 13:R2

Immunohistochemical analysis of Human salivary glands taken from
patients with primary Sjögren's syndrome, staining Bcl-2 with
unpurified ab32124.

Antigen retrieval was performed via heat mediation in a citrate
buffer (pH 6). Sections were blocked using 2% BSA, 10% normal
serum and permeabilized with 0.5% Triton X-100. Samples were
incubated with primary antibody (1/100) for one hour at room
temperature. An Alexa Fluor® 594-conjugated anti-rabbit IgG was
used as the secondary antibody.

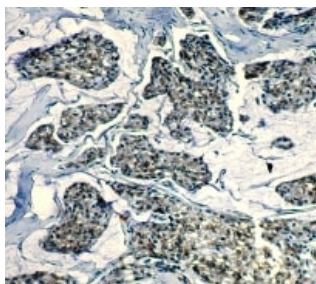
N.B. Panels B and D are higher magnifications of panels A and C,
respectively.



OIR-D Scanning - Anti-Bcl-2 antibody [E17]
(ab32124)

Equilibrium disassociation constant (K_D)

[Click here to learn more about \$K_D\$](#)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bcl-2 antibody [E17] (ab32124)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast carcinoma tissue labelling Bcl-2 with unpurified ab32124 at 1/200 dilution. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.

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Recombinant technology



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Anti-Bcl-2 antibody [E17] (ab32124)

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