

Anti-Bad antibody [Y208] - BSA and Azide free ab220116

敲除验证
重组
RabMAb

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概述

产品名称	Anti-Bad抗体[Y208] - BSA and Azide free
描述	兔单克隆抗体[Y208] to Bad - BSA and Azide free
宿主	Rabbit
特异性	<p>This antibody does not cross-react with other Bcl-2 members.</p> <p>The mouse and rat recommendation is based on the IHC-P results. We do not guarantee WB for mouse and rat.</p>
经测试应用	适用于: Flow Cyt (Intra), ICC/IF, WB, IHC-P
种属反应性	<p>与反应: Mouse, Rat, Human</p> <p>预测可用于: Dog </p>
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	IHC-P: Human colon, ovarian cancer, Mouse and Rat kidney tissue; ICC/IF: HeLa cells; Flow Cyt (intra): MCF7 cells. WB: HeLa and HepG2 whole cell lysate.
常规说明	<p>ab220116 is the carrier-free version of ab32445.</p> <p>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.</p> <p>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.</p> <p>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.</p> <p>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.</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

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 Constituent: PBS
无载体	是
纯度	Protein A purified
克隆	单克隆
克隆编号	Y208
同种型	IgG

应用

The Abpromise guarantee

Abpromise[™]承诺保证使用ab220116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。

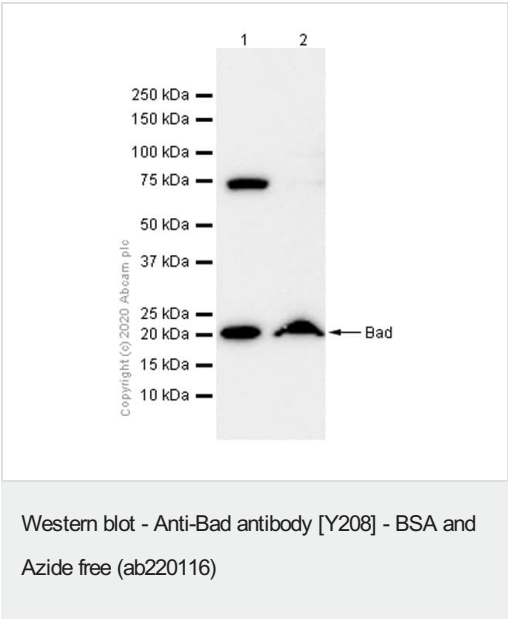
应用	Ab评论	说明
Flow Cyt (Intra)		Use at an assay dependent concentration. ab199376 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 23 kDa (predicted molecular weight: 18 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. The mouse and rat recommendation is based on the IHC-P results. We do not guarantee WB for mouse and rat.

靶标

功能	Promotes cell death. Successfully competes for the binding to Bcl-X(L), Bcl-2 and Bcl-W, thereby affecting the level of heterodimerization of these proteins with BAX. Can reverse the death repressor activity of Bcl-X(L), but not that of Bcl-2 (By similarity). Appears to act as a link between growth factor receptor signaling and the apoptotic pathways.
组织特异性	Expressed in a wide variety of tissues.

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.
翻译后修饰	Phosphorylated on one or more of Ser-75, Ser-99, Ser-118 and Ser-134 in response to survival stimuli, which blocks its pro-apoptotic activity. Phosphorylation on Ser-99 or Ser-75 promotes heterodimerization with 14-3-3 proteins. This interaction then facilitates the phosphorylation at Ser-118, a site within the BH3 motif, leading to the release of Bcl-X(L) and the promotion of cell survival. Ser-99 is the major site of AKT/PKB phosphorylation, Ser-118 the major site of protein kinase A (CAPK) phosphorylation. Ser-75 is phosphorylated by AKT/PKB, protein kinase A and PIM2.
细胞定位	Mitochondrion outer membrane. Cytoplasm. Upon phosphorylation, locates to the cytoplasm.

图片



All lanes : Anti-Bad antibody [Y208] ([ab32445](#)) at 1/2000 dilution (Purified)

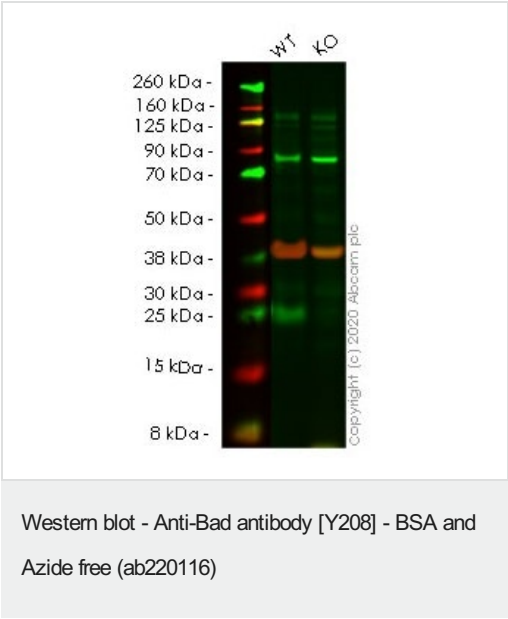
Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 2 : HepG2 (Human hepatocellular carcinoma epithelial cell) whole cell lysate

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 18 kDa



All lanes : Anti-Bad antibody [Y208] ([ab32445](#)) at 1/2000 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : BAD knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 18 kDa

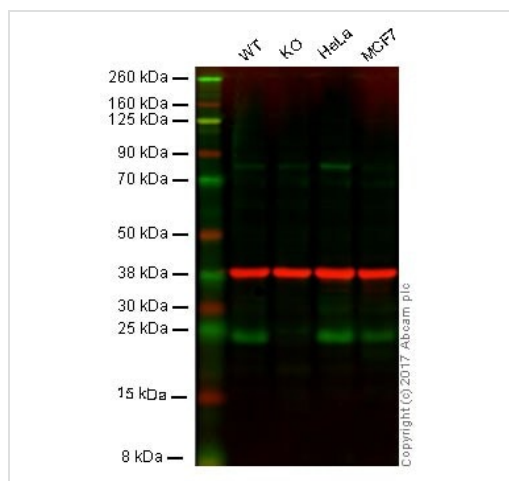
Observed band size: 23 kDa

This data was developed using the same antibody clone in a

different buffer formulation ([ab32445](#)).

Lanes 1- 2: Merged signal (red and green). Green - [ab32445](#) observed at 23 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) observed at 37 kDa.

[ab32445](#) was shown to react with Bad in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line [ab264843](#) (knockout cell lysate [ab256847](#)) was used. Wild-type HeLa and BAD 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. [ab32445](#) 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.



Western blot - Anti-Bad antibody [Y208] - BSA and Azide free ([ab220116](#))

All lanes : Anti-Bad antibody [Y208] ([ab32445](#)) at 1/2000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate

Lane 2 : BAD knockout HAP1 whole cell lysate

Lane 3 : HeLa whole cell lysate

Lane 4 : MCF7 whole cell lysate

Lysates/proteins at 20 µg per lane.

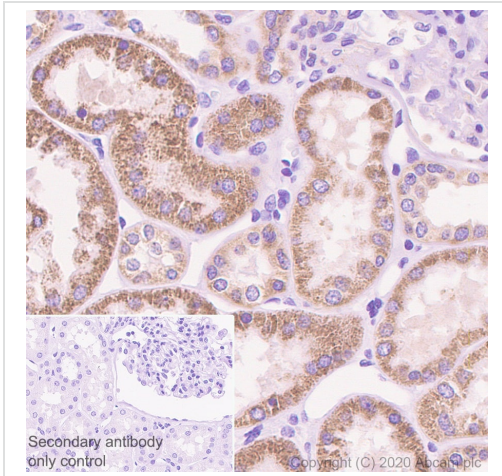
Predicted band size: 18 kDa

This data was developed using the same antibody clone in a different buffer formulation ([ab32445](#)).

Lanes 1 - 4: Merged signal (red and green). Green - [ab32445](#) observed at 23 kDa. Red - loading control, [ab9484](#), observed at 37 kDa.

[ab32445](#) was shown to specifically recognise BAD in wild-type HAP1 cells along with additional cross reactive bands. No band was observed when BAD knockout cells were examined. Wild-type and BAD knockout samples were subjected to SDS-PAGE. Ab32445 and [ab9484](#) (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/2000 dilution and 1/20,000 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 1/20,000 dilution for 1 hour at room temperature before imaging.

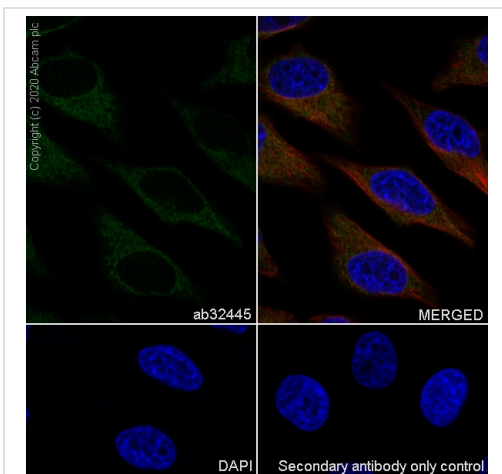


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bad antibody [Y208] - BSA and Azide free (ab220116)

This data was developed using **ab32445**, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human kidney tissue sections labeling Bad with purified **ab32445** at 1/1000 dilution (0.14 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) secondary antibody was used at 1/0 dilution. PBS instead of the primary antibody was used as the negative control.

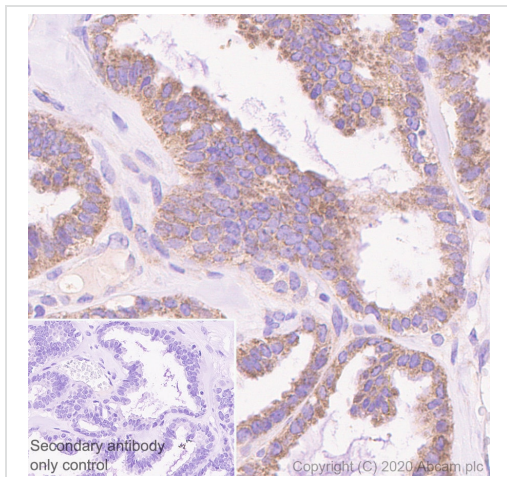
The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Immunocytochemistry/ Immunofluorescence - Anti-Bad antibody [Y208] - BSA and Azide free (ab220116)

This data was developed using **ab32445**, the same antibody clone in a different buffer formulation.

Immunocytochemistry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Bad with purified **ab32445** at 1/50 dilution (2.9 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-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 IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

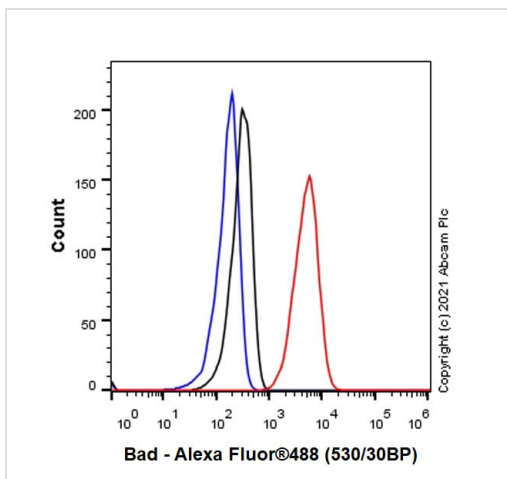


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bad antibody [Y208] - BSA and Azide free (ab220116)

This data was developed using [ab32445](#), the same antibody clone in a different buffer formulation.

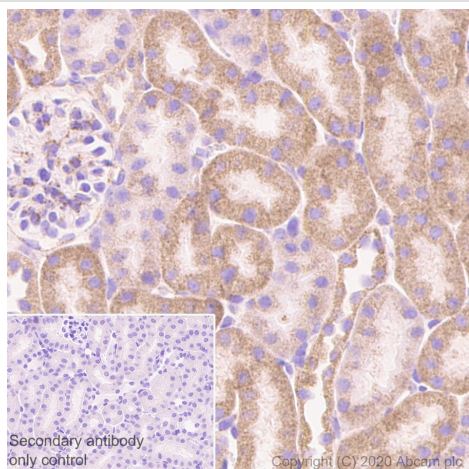
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human ovarian cancer tissue sections labeling Bad with purified [ab32445](#) at 1/1000 dilution (0.14 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)) secondary antibody was used at 1/0 dilution. PBS instead of the primary antibody was used as the negative control.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Flow Cytometry (Intracellular) - Anti-Bad antibody [Y208] - BSA and Azide free (ab220116)

This data was developed using [ab32445](#), the same antibody clone in a different buffer formulation. Intracellular Flow Cytometry analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labelling Bad with purified [ab32445](#) at 1/20 dilution (10 µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, [ab150077](#)) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).

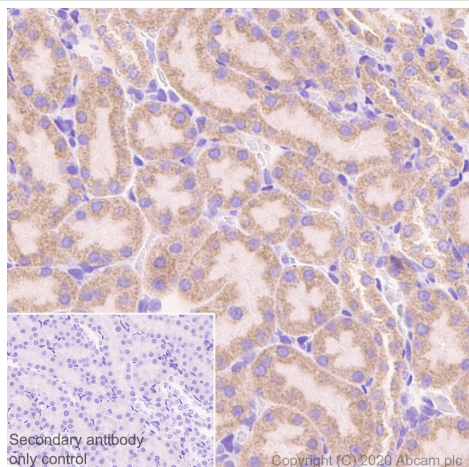


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bad antibody [Y208] - BSA and Azide free (ab220116)

This data was developed using [ab32445](#), the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue sections labeling Bad with purified [ab32445](#) at 1/1000 dilution (0.14 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)) secondary antibody was used at 1/0 dilution. PBS instead of the primary antibody was used as the negative control.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Bad antibody [Y208] - BSA and Azide free (ab220116)

This data was developed using [ab32445](#), the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat kidney tissue sections labeling Bad with purified [ab32445](#) at 1/1000 dilution (0.14 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)) secondary antibody was used at 1/0 dilution. PBS instead of the primary antibody was used as the negative control.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-Bad antibody [Y208] - BSA and Azide free
(ab220116)

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