# abcam

## Product datasheet

## Anti-Survivin antibody [EP2880Y] ab76424

Recombinant RabMAb

★★★★★ 9 Abreviews 147 References 20 Images

#### Overview

**Product name** Anti-Survivin antibody [EP2880Y]

**Description** Rabbit monoclonal [EP2880Y] to Survivin

**Host species** Rabbit

**Tested applications** Suitable for: IHC-P, WB, IP, ICC/IF, Sandwich ELISA, Flow Cyt (Intra)

Species reactivity Reacts with: Human

Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. **Immunogen** 

Positive control HeLa and Jurkat lysates; human urinary bladder carcinoma and human colonic adenocarcinoma

tissues; A549; Ramos; Human bladder carcinoma flow cyt (intra): Human PBMCs

**General notes** 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**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

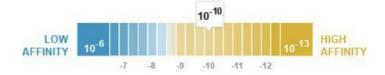
### **Properties**

**Form** Liquid

Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long Storage instructions

term. Avoid freeze / thaw cycle.

 $K_D = 4.40 \times 10^{-10} M$ Dissociation constant (K<sub>D</sub>)



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

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.5% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EP2880Y

**Isotype** IgG

#### **Applications**

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab76424 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P	<b>★★★★★</b> (2)	1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Signal amplification system might be necessary or the use of polymerized HRP secondary antibodies.  For unpurified use at 1/250 - 1/500.
WB	★★★★☆(3)	1/5000 - 1/20000. Detects a band of approximately 16 kDa (predicted molecular weight: 16 kDa).
IP		1/150. For unpurified use at 1:10.
ICC/IF	*** <u>*</u>	1/100 - 1/250.
Sandwich ELISA		Use a concentration of 0.5 $\mu$ g/ml. For sandwich ELISA, use this antibody as detection at 0.5 $\mu$ g/ml with <u>ab27468</u> as capture.
Flow Cyt (Intra)		1/500.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody. For unpurified use at 1/50.

#### **Target**

#### **Function**

Component of the chromosomal passenger complex (CPC), a complex that acts as a key regulator of mitosis. The CPC complex has essential functions at the centromere in ensuring correct chromosome alignment and segregation and is required for chromatin-induced microtubule stabilization and spindle assembly. The complex with RAN plays a role in mitotic spindle formation by serving as a physical scaffold to help deliver the RAN effector molecule TPX2 to microtubules. May play a role in neoplasia. May counteract a default induction of apoptosis in G2/M phase. Inhibitor of caspase-3 and caspase-7. Isoform 2 and isoform 3 do not appear to play vital roles in mitosis. Isoform 3 shows a marked reduction in its anti-apoptotic effects when compared with the displayed wild-type isoform.

**Tissue specificity** Expressed only in fetal kidney and liver, and to lesser extent, lung and brain. Abundantly

expressed in adenocarcinoma (lung, pancreas, colon, breast, and prostate) and in high-grade

lymphomas. Also expressed in various renal cell carcinoma cell lines.

**Sequence similarities**Belongs to the IAP family.

Contains 1 BIR repeat.

**Developmental stage** Expression is cell cycle-dependent and peaks at mitosis.

**Domain** The BIR repeat is necessary and sufficient for HBXIP binding.

**Post-translational** Ubiquitination is required for centrosomal targeting.

modifications In vitro phosphorylation at Thr-117 by AURKB/STK12 prevents interaction with INCENP and

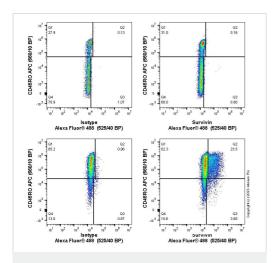
localization to mitotic chromosomes.

Cytoplasm. Nucleus. Chromosome > centromere. Cytoplasm > cytoskeleton >

spindle. Localizes on chromosome arms and inner centromeres from prophase through metaphase and then transferring to the spindle midzone and midbody from anaphase through

cytokinesis. Colocalizes with AURKB at mitotic chromosomes.

#### **Images**

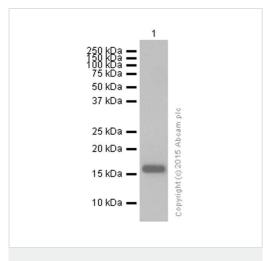


Flow Cytometry (Intracellular) - Anti-Survivin antibody [EP2880Y] (ab76424)

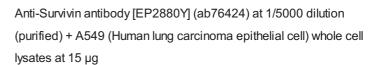
Flow cytometry staining of human peripheral blood mononuclear cells (PBMCs) (top) or PBMCs treated with 1µg/mL PHA for 24h followed by 20U/mL IL-2 for 48h (bottom), with ab76424 (right) or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (left). Cells were fixed and permeabilised with BD Cytofix/Cytoperm for 20 min. PBMCs were incubated for 30 min at 4°C in 1x PBS containing 10 µg/ml human IgG and 10 % normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody ab76424 or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (1x 10 $^6$  in 100 µl at 0.2 µg/ml (1/11100)) for 30 min at 4°C . The cells were simultaneously stained with CD45RO.

The secondary antibody Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 4°C

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on CD4+ cells.



Western blot - Anti-Survivin antibody [EP2880Y] (ab76424)

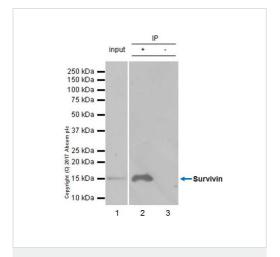


#### Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 16 kDa

Blocking and diluting buffer: 5% NFDM/TBST



Immunoprecipitation - Anti-Survivin antibody [EP2880Y] (ab76424)

ab76424 (purified) at 1:150 dilution (5 $\mu$ g) immunoprecipitating Survivin in Ramos whole cell lysate.

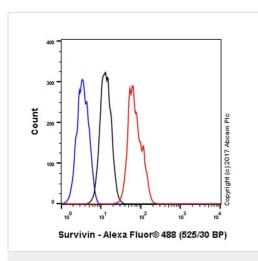
Lane 1 (input): Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysate 10µg

Lane 2 (+): ab76424 & Ramos whole cell lysate

Lane 3 (-): Rabbit monoclonal  $\lg G$  ( $\underline{ab172730}$ ) instead of ab76424 in Ramos whole cell lysate

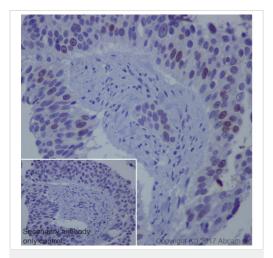
For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1:1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST.



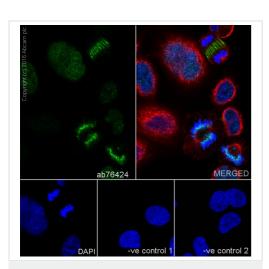
Flow Cytometry (Intracellular) - Anti-Survivin antibody [EP2880Y] (ab76424)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Survivin with purified ab76424 at 1/500 dilution (5  $\mu$ g/ml) (red). Cells were fixed with 80% methanol. A Goat anti rabbit lgG (Alexa Fluor 488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

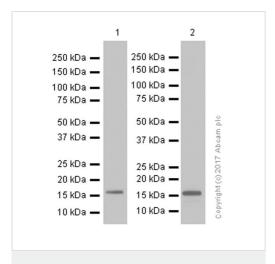


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human bladder carcinoma tissue sections labeling Survivin with Purified ab76424 at 1:1000 dilution (2.52 µg/ml). Heat mediated antigen retrieval was performed using Tris/EDTA Buffer, pH 9.0. Tissue was counterstained with Hematoxylin. <a href="mailto:ab97051">ab97051</a> Goat Anti-Rabbit lgG H&L (HRP) secondary antibody was used at 1:500 dilution. PBS instead of the primary antibody was used as the negative control.



Immunocytochemistry/ Immunofluorescence - Anti-Survivin antibody [EP2880Y] (ab76424)



Western blot - Anti-Survivin antibody [EP2880Y] (ab76424)

Immunofluorescent staining of MCF-7 cells labelling Bcl-2 with purified ab76424 at 1/1000. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. The cells were costained with **ab7291**, a mouse anti-tubulin (1/1000) using **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/1000) as the secondary antibody. Nuclei counterstained with DAPI (blue).

Control 1: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).

Control 2: <u>ab7291</u> (1/1000) and secondary antibody, <u>ab150077</u>, an Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit lgG (1/500).

**All lanes :** Anti-Survivin antibody [EP2880Y] (ab76424) at 1/5000 dilution (purified)

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

Lane 2 : Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysates

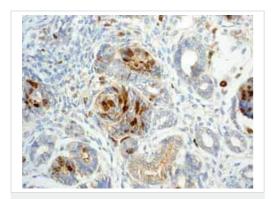
Lysates/proteins at 15 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 16 kDa

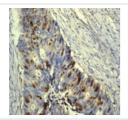
Blocking and diluting buffer: 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

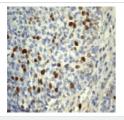
Unpurified ab76424 showing positive staining in Fetal kidney tissue. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.

Flow Cytometry (Intracellular) - Anti-Survivin antibody [EP2880Y] (ab76424) Overlay histogram showing HeLa cells stained with unpurified ab76424 (red line). The cells were fixed with 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 (ab76424, 1/50 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit monoclonal lgG (1µg/1x106 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a decreased signal in HeLa cells fixed with 4% paraformaldehyde/permeabilized with 0.1% PBS-Tween 20 used under the same conditions.



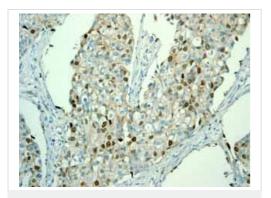
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

Unpurified ab76424 at 1/250 dilution staining Survivin in human colonic adenocarcinoma by Immunohistochemistry, Paraffinembedded tissue. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



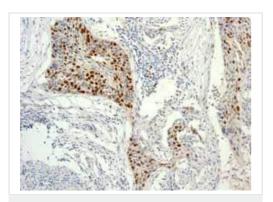
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

Unpurified ab76424 at 1/250 dilution staining Survivin in human urinary bladder carcinoma by Immunohistochemistry, Paraffinembedded tissue. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



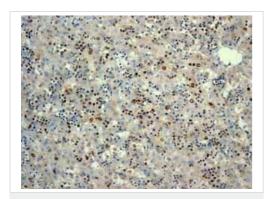
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

Unpurified ab76424 showing positive staining in breast carcinoma tissue. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



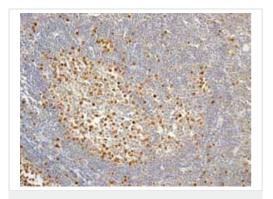
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

Unpurified ab76424 showing positive staining in cervical carcinoma tissue. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



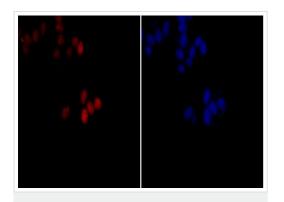
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

Unpurified ab76424 showing positive staining in fetal liver tissue. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



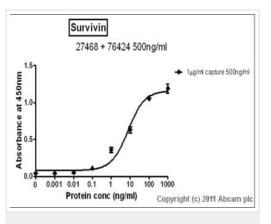
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Survivin antibody
[EP2880Y] (ab76424)

Unpurified ab76424 showing positive staining in normal tonsil tissue. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Survivin antibody [EP2880Y] (ab76424)

Immunocytochemistry/Immunofluorescence analysis of MCF-7 cells labelling Survivin with unpurified ab76424 at 1/100. Cells were fixed with 4% paraformaldehyde for 20 minutes at room temperature. Cells were incubated with the primary antibody overnight. An Alexa Fluor<sup>®</sup> 555-conjugated anti-rabbit lgG was used as the secondary antibody. Left - Survivin, Right - DAPI.



Sandwich ELISA - Anti-Survivin antibody [EP2880Y] (ab76424)

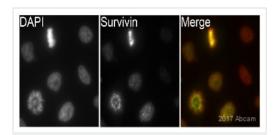
Standard curve for Survivin (Analyte: <u>ab87202</u>); dilution range 1pg/ml to 1µg/ml using Capture Antibody <u>ab27468</u> at 1µg/ml and Detector Antibody abRabbit monoclonal [EP2880Y] to Survivin (ab76424) at 0.5µg/ml. Concentration of Rabbit monoclonal [EP2880Y] to Survivin (ab76424) may vary from lot to lot; please use this curve as guideline.

Ol-RD Scanning - Anti-Survivin antibody [EP2880Y] (ab76424)

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

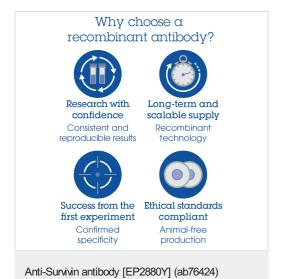
## Click here to learn more about K<sub>D</sub>

ab76424 staining Survivin in the Hela cell line from Human cervix by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde, permeabilized with 0.5% Triton X-100. Samples were incubated with primary antibody (1/500 in PBS) for 1 hour at 22°C. Ab150081 (1/200) was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-Survivin antibody [EP2880Y] (ab76424)

This image is courtesy of an abreview by Kirk Mcmanus



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