abcam

Product datasheet

Anti-MCL1 antibody [Y37] - BSA and Azide free ab186822

敲除验证 重组 RabMAb

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概述

产品名称	Anti-MCL1抗体[Y37] - BSA and Azide free
描述	兔单克隆抗体[Y37] to MCL1 - BSA and Azide free
宿主	Rabbit
特异性	This antibody recognises MCL1. The antibody does not cross-react with other Bcl-2 family members.
经测试应用	**适用于:** Flow Cyt (Intra), IHC-P, WB, ICC/IF
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: HEK293T and Ramos cell lysates. IHC-P: Human colon adenocarcinoma tissue. Flow Cyt (intra): Ramos and A431 cells. ICC/IF: HCT116 and H1299 cells.
常规说明	ab186822 is the carrier-free version of **ab32087**. 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. 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. 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. 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. 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**.

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	Y37
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab186822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。

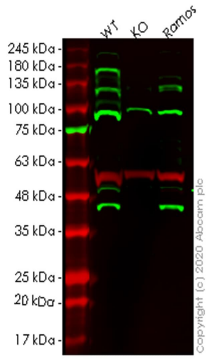
应用	Ab评论	说明
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.
WB		Use at an assay dependent concentration. Predicted molecular weight: 37 kDa. Can be blocked with MCL1 peptide (**ab199979**).
ICC/IF	★☆☆☆☆ **(1)**	Use at an assay dependent concentration.

靶标

功能	Involved in the regulation of apoptosis versus cell survival, and in the maintenance of viability but not of proliferation. Mediates its effects by interactions with a number of other regulators of apoptosis. Isoform 1 inhibits apoptosis. Isoform 2 promotes apoptosis.
序列相似性	Belongs to the Bcl-2 family.
翻译后修饰	Cleaved by CASP3 during apoptosis. In intact cells cleavage occurs preferentially after Asp-127, yielding a pro-apoptotic 28 kDa C-terminal fragment. Rapidly degraded in the absence of phosphorylation on Thr-163 in the PEST region. Phosphorylated on Thr-163. Treatment with taxol or okadaic acid induces phosphorylation on additional sites.

细胞定位	Membrane. Cytoplasm. Mitochondrion. Nucleus > nucleoplasm. Cytoplasmic, associated with mitochondria.

图片



Western blot - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

All lanes : Anti-MCL1 antibody [Y37] (**ab32087**) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : MCL1 knockout HEK293T cell lysate
Lane 3 : Ramos cell lysate

Lysates/proteins at 20 µg per lane.

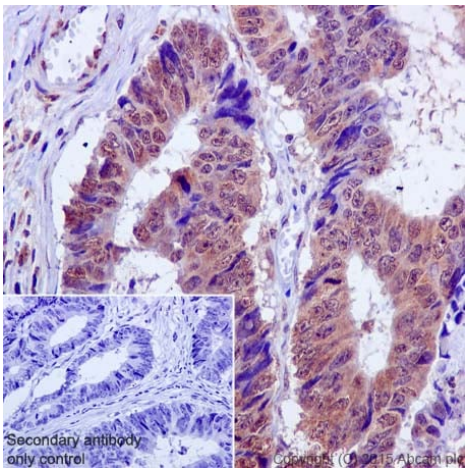
Secondary
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

Predicted band size: 37 kDa
Observed band size: 37 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab32087**).

Lanes 1-3: Merged signal (red and green). Green - **ab32087** observed at 37 kDa. Red - loading control **ab7291** observed at 50 kDa.

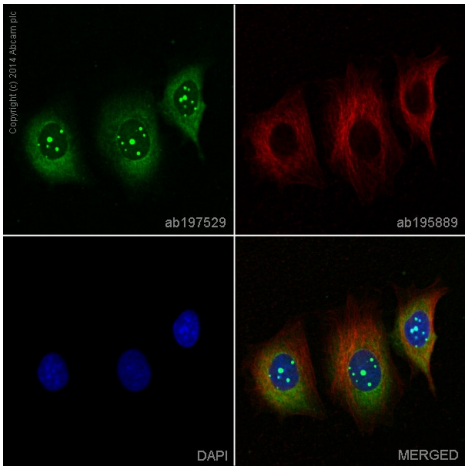
ab32087 Anti-MCL1 antibody [Y37] was shown to specifically react with MCL1 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line **ab266838** (knockout cell lysate **ab256986**) was used. Wild-type and MCL1 knockout samples were subjected to SDS-PAGE. **ab32087** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) were incubated overnight at 4°C at 1 in 1000 dilution and 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-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

This IHC data was generated using the same anti-MCL1 antibody clone, Y37, in a different buffer formulation (cat# **ab32087**).

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon adenocarcinoma tissue labelling MCL1 with purified **ab32087** at 1/100. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a goat anti-rabbit IgG H&L (HRP) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

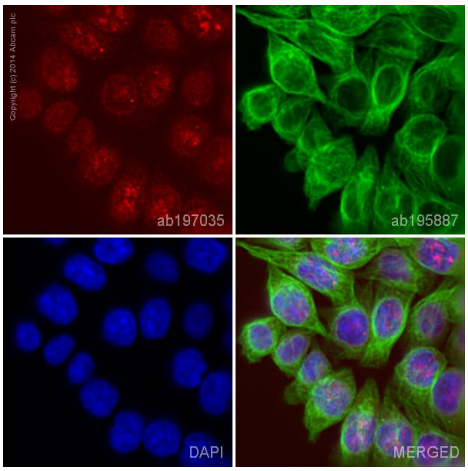


Immunocytochemistry/ Immunofluorescence - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

Clone Y37 (ab186822) has been successfully conjugated by Abcam. This image was generated using Anti-MCL1 antibody [Y37] (Alexa Fluor® 488). Please refer to **ab197529** for protocol details.

ab197529 staining MCL1 in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilised in 0.1% Triton X-100 for 5 minutes and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab197529** at 1/100 dilution (shown in green) and **ab195889**, Mouse monoclonal [DM1A] to alpha Tubulin (Alexa Fluor® 594, shown in red) at 1/167 dilution overnight at +4°C. Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

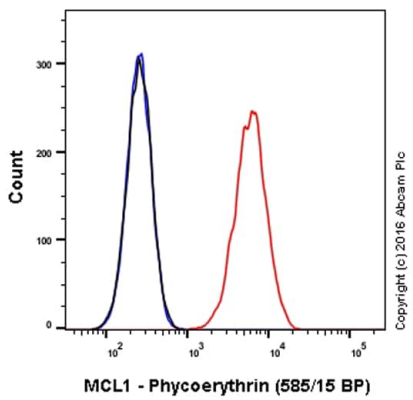


Immunocytochemistry/ Immunofluorescence - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

Clone Y37 (ab186822) has been successfully conjugated by Abcam. This image was generated using Anti-MCL1 antibody [Y37] (Alexa Fluor® 647). Please refer to **ab197035** for protocol details.

ab197035 staining MCL1 in HCT116 cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab at a 1/100 dilution (shown in red) and **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 488), at a 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



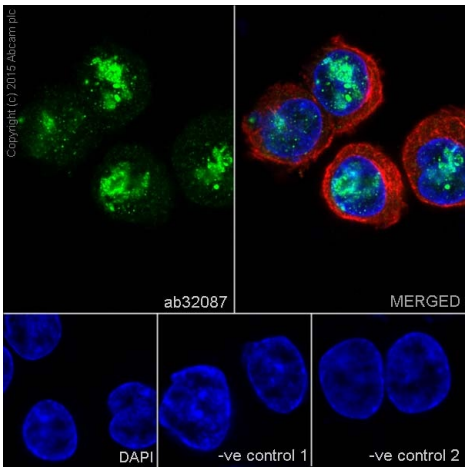
Flow Cytometry (Intracellular) - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

Clone Y37 (ab186822) has been successfully conjugated by Abcam. This image was generated using Anti-MCL1 antibody [Y37] (PE). Please refer to **ab209289** for protocol details.

Overlay histogram showing MCF7 cells stained with **ab209289** (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 90% methanol for 30 min at -20°C. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (**ab209289**, 1/2500 dilution) for 30 min at 22°C.

Isotype control antibody (black line) was rabbit IgG (monoclonal) Phycoerythrin (**ab209478**) used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5,000 events were collected using a 50 mW Yellow/Green laser (561nm) and 586/15 bandpass filter.



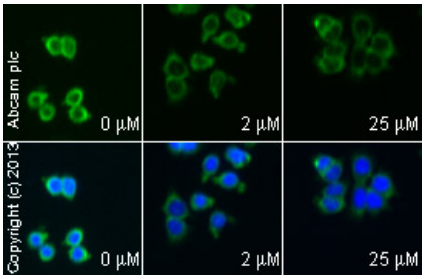
Immunocytochemistry/ Immunofluorescence - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

Immunocytochemistry/Immunofluorescence analysis of HCT 116 (human colorectal carcinoma cell line) cells labelling MCL1 with purified **ab32087** at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. **ab7291**, a mouse anti-tubulin (1/1000) and **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000) were also used.

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

Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).

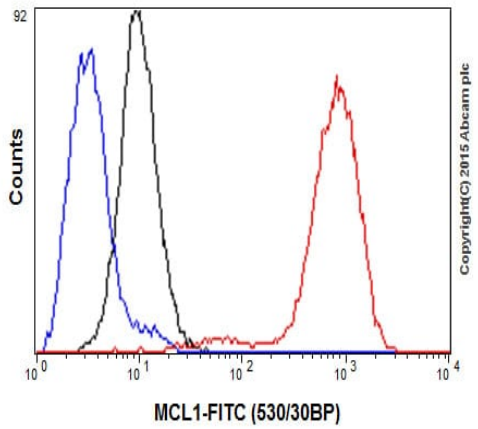
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32087**).



Immunocytochemistry/ Immunofluorescence - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

Immunocytochemistry/Immunofluorescence analysis of HCT 116 (human colorectal carcinoma cell line) cells treated with wogonin (**ab142471**) labelling MCL1 with unpurified **ab32087**. Decrease of MCL1 expression correlates with increased concentration of wogonin, as described in literature. Cells were incubated at 37°C for 2h in media containing different concentrations of **ab142471** (wogonin) in DMSO, fixed with 4% formaldehyde for 10 minutes at room temperature and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with **ab32087** (1/100) dilution was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight 488 anti-rabbit polyclonal antibody (**ab96899**) at 1/250 dilution was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

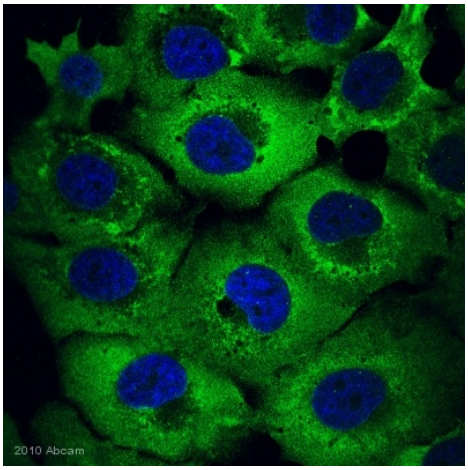
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32087**).



Flow Cytometry (Intracellular) - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

Intracellular Flow Cytometry analysis of Ramos (human Burkitt's lymphoma cell line) cells labelling MCL1 with purified **ab32087** at 1/250 (red). Cells were fixed with 4% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32087**).

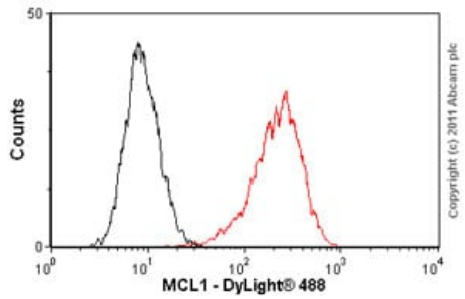


Immunocytochemistry/ Immunofluorescence - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

This image is courtesy of an anonymous Abreview.

Immunocytochemistry/Immunofluorescence analysis of H1299 cells labelling MCL1 with unpurified **ab32087**. Cells were PFA-fixed and permeabilized in 0.5% Triton X-100 prior to blocking in 3% Serum for 1 hour at 24°C. The primary antibody was diluted 1/100 and incubated with the sample for 1 hour at 24°C. The secondary antibody was an Alexa Fluor® 488-conjugated Goat anti-Rabbit polyclonal, diluted 1/2000. DAPI (blue) was used as the nuclear counterstain.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32087**).



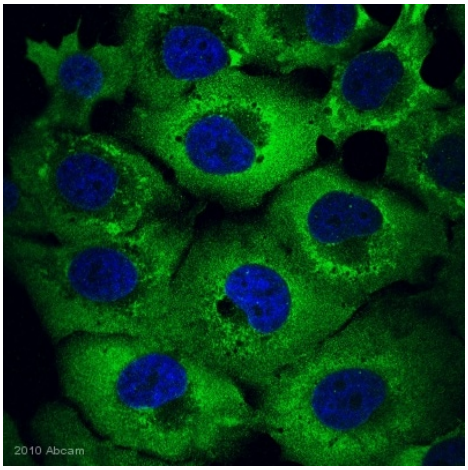
Flow Cytometry (Intracellular) - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

Intracellular Flow Cytometry analysis of A431 (human epidermoid carcinoma cell line) cells labelling MCL1 with unpurified **ab32087** (red line). 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 (**ab32087**, 1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (**ab96899**) at 1/500 dilution for 30 min at 22°C.

Black - Isotype control, rabbit monoclonal IgG.

Acquisition of >5,000 events was performed. This antibody gave a decreased signal in A431 cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween used under the same conditions.

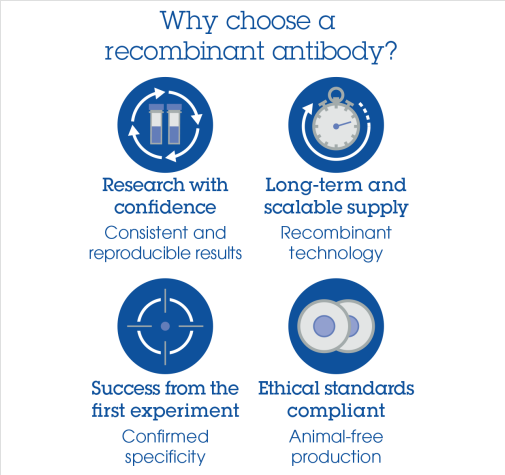
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32087**).



Immunocytochemistry/ Immunofluorescence - Anti-MCL1 antibody [Y37] - BSA and Azide free (ab186822)

This ICC/IF data was generated using the same anti-MCL1 antibody clone, Y37, in a different buffer formulation (cat# **ab32087**).

Immunocytochemistry/Immunofluorescence analysis of H1299 cells labelling MCL1 with unpurified **ab32087**. Cells were PFA-fixed and permeabilized in 0.5% Triton X-100 prior to blocking in 3% Serum for 1 hour at 24°C. The primary antibody was diluted 1/100 and incubated with the sample for 1 hour at 24°C. The secondary antibody was an Alexa Fluor® 488-conjugated Goat anti-Rabbit polyclonal, diluted 1/2000. DAPI (blue) was used as the nuclear counterstain.



Anti-MCL1 antibody [Y37] - BSA and Azide free

(ab186822)

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