abcam

Product datasheet

Anti-Mark3 antibody [EPR633Y] ab52626

敲除验证 重组 RabMAb

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概述

产品名称	Anti-Mark3抗体[EPR633Y]
描述	兔单克隆抗体[EPR633Y] to Mark3
经测试应用	**适用于:** WB, IP, Flow Cyt, ICC/IF **不适用于:** IHC-P
种属反应性	**与反应:** Mouse, Human
免疫原	Synthetic peptide (the amino acid sequence is considered to be commercially sensitive) within Human Mark3 aa 600-700 (C terminal). The exact sequence is proprietary.
阳性对照	WB: HeLa, K562 and NIH/3T3 cell lysate. ICC/IF: MCF-7 cells. Flow Cyt: HeLa cells. IP: K562 cells.
常规说明	This product is a recombinant rabbit monoclonal antibody. Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab® patents **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.**

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.
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	EPR633Y
同种型	IgG

应用

Our Abpromise guarantee covers the use of **ab52626** in the following tested applications.

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

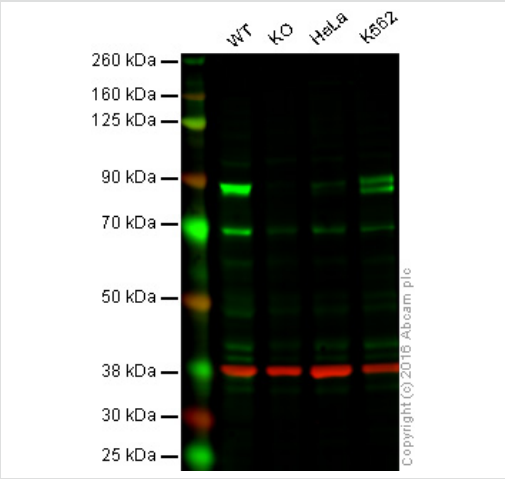
应用	Ab评论	说明
WB		1/1000 - 1/2000. Detects a band of approximately 86 kDa.
IP	★★★★★	1/20 - 1/60.
Flow Cyt		1/30 - 1/1000. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/50 - 1/100.

应用说明	Is unsuitable for IHC-P.

靶标

功能	Involved in the specific phosphorylation of microtubule-associated proteins for tau, MAP2 and MAP4. Phosphorylates CDC25C on 'Ser-216'.
组织特异性	Ubiquitous.
序列相似性	Belongs to the protein kinase superfamily. CAMK Ser/Thr protein kinase family. MARK subfamily. Contains 1 KA1 (kinase-associated) domain. Contains 1 protein kinase domain. Contains 1 UBA domain.

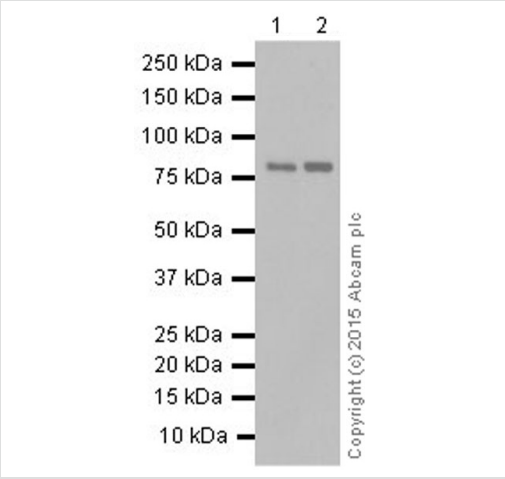
图片



Western blot - Anti-Mark3 antibody [EPR633Y] (ab52626)

Lane 1: Wild-type HAP1 cell lysate (40 µg)
Lane 2: MARK3 knockout HAP1 cell lysate (40 µg)
Lane 3: HeLa cell lysate (20 µg)
Lane 4: K562 cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab52626 observed at 85 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab52626 was shown to recognize Mark3 when Mark3 knockout samples were used, along with additional cross-reactive bands. Wild-type and Mark3 knockout samples were subjected to SDS-PAGE. Ab52626 and ab8245 (loading control to GAPDH) were diluted at 1/1000 and 1:10,000 dilution respectively and incubated overnight at 4C. 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 1:10,000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Mark3 antibody [EPR633Y] (ab52626)

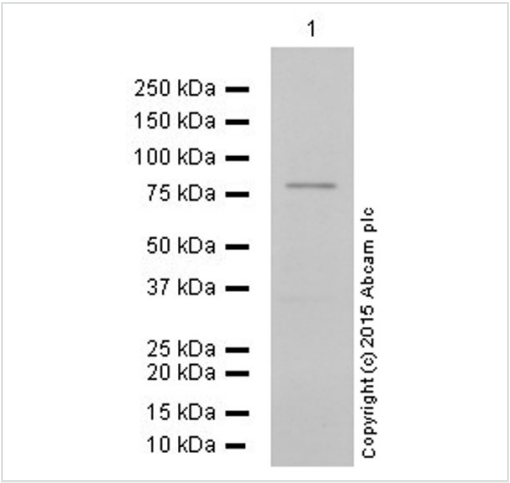
All lanes : Anti-Mark3 antibody [EPR633Y] (ab52626) at 1/2000 dilution (purified)

Lane 1 : K562 whole cell lysate
Lane 2 : HeLa whole cell lysate

Lysates/proteins at 20 µg per lane.

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

Observed band size : 86 kDa
Blocking and dilution buffer: 5% NFDM/TBST



Western blot - Anti-Mark3 antibody [EPR633Y] (ab52626)

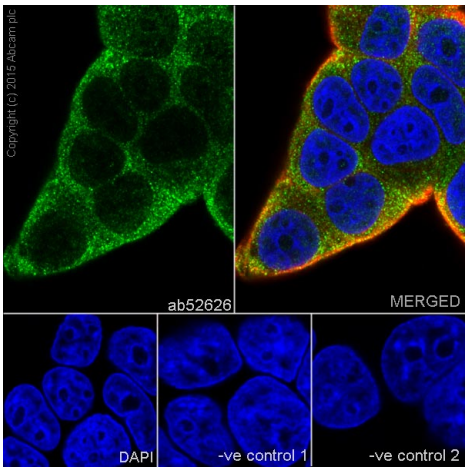
Anti-Mark3 antibody [EPR633Y] (ab52626) at 10 µg (purified) + NIH/3T3 whole cell lysate at 10 µg

Secondary

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

Observed band size : 86 kDa

Blocking and dilution buffer: 5% NFDM/TBST

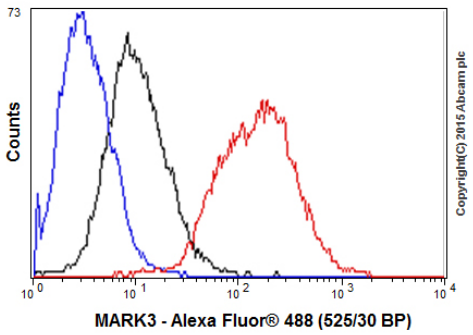


Immunocytochemistry/ Immunofluorescence - Anti-Mark3 antibody [EPR633Y] (ab52626)

Immunocytochemistry/Immunofluorescence analysis of MCF-7 cells labelling Mark3 with purified ab52626 at a dilution of 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) 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.

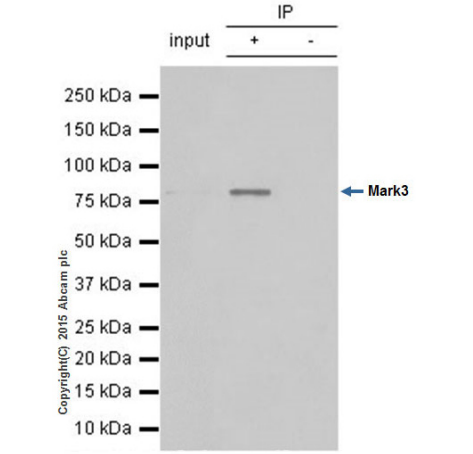
Control 1: primary antibody (1/100) and secondary antibody, ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000).

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



Flow Cytometry - Anti-Mark3 antibody [EPR633Y] (ab52626)

Flow Cytometry analysis of HeLa cells labelling Mark3 with purified ab52626 at a dilution of 1/50 (red). Cells were fixed with 4% paraformaldehyde. An Alexa Fluor® 488-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.



Immunoprecipitation - Anti-Mark3 antibody [EPR633Y] (ab52626)

ab52626 (purified) at a dilution of 1/20 immunoprecipitating Mark3 in K562 whole cell lysate.

Lane 1 (input): K562 whole cell lysate (10µg)

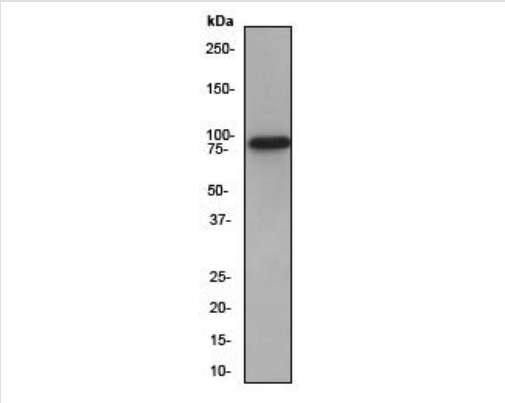
Lane 2 (+): ab52626 + K562 whole cell lysate.

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab52626 in K562 whole cell lysate.

For western blotting, ab131366 VeriBlot for IP (HRP) was used as the secondary antibody (1/1000).

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



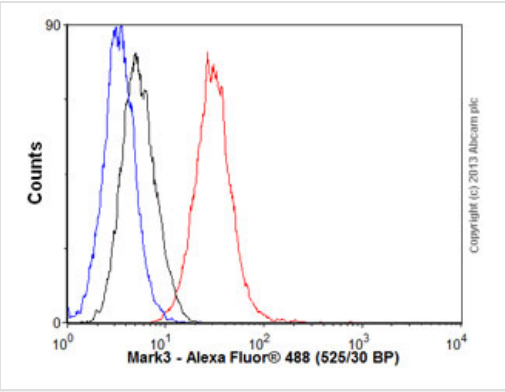
Western blot - Anti-Mark3 antibody [EPR633Y] (ab52626)

Anti-Mark3 antibody [EPR633Y] (ab52626) at 1/2000 dilution (unpurified) + HeLa cell lysate at 10 µg

Secondary

HRP-conjugated goat anti-rabbit IgG at 1/2000 dilution

Observed band size : 86 kDa



Flow Cytometry - Anti-Mark3 antibody [EPR633Y] (ab52626)

Overlay histogram showing HeLa cells stained with unpurified ab52626 (red line). The cells were fixed with 80% 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 (unpurified ab52626, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

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