abcam

Product datasheet

Anti-Mark3 antibody ab77199

6 图像

概述

产品名称	Anti-Mark3抗体
描述	**小鼠单克隆抗体**to Mark3
宿主	Mouse
经测试应用	**适用于:** WB, ICC/IF, Flow Cyt, ELISA
种属反应性	**与反应:** Human **预测可用于:** Mouse, Rat, Dog, Chimpanzee
免疫原	Recombinant fragment: HHKVQRSVFS SQKQRRYSDH AGPAIPSVVA YPKRSQTSTA DSDLKEDGIS SRKSSGSAVG GKGIAPASPM LGNASNPNKA DIPERKKSST VPSSNTASG, corresponding to amino acids 402-500 of Human Mark3 (NP_002367) with a 26 kDa tag. Run BLAST with ExPASy Run BLAST with NCBI
阳性对照	Mark3 transfected 293T cell lysate. Recombinant tagged human Mark3 fragment.

性能

形式	Liquid
存放说明	Shipped at 4°C. Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
存储溶液	Preservative: None Constituents: 1X PBS, pH 7.2
纯度	Protein A purified
克隆	单克隆
同种型	IgG2b
轻链类型	kappa

应用

Our Abpromise guarantee covers the use of **ab77199** in the following tested applications.

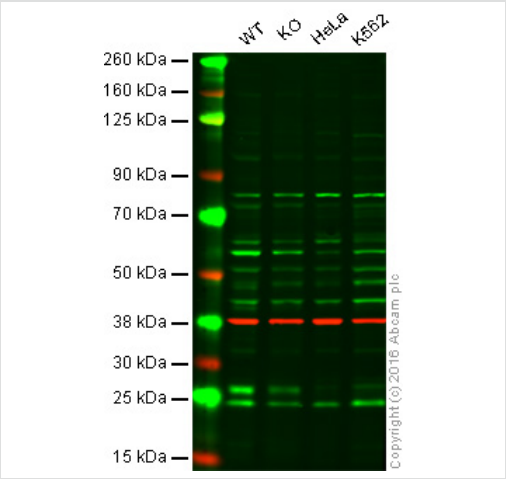
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

应用	Ab评论	说明
WB		Use a concentration of 1 - 5 µg/ml. Predicted molecular weight: 87 kDa.
ICC/IF		Use a concentration of 5 µg/ml.
Flow Cyt		Use 0.01-0.1µg for 10^6 cells. ab170192 - Mouse monoclonal IgG2b, is suitable for use as an isotype control with this antibody.
ELISA		Use at an assay dependent concentration. Detection limit for recombinant tagged Mark3 is approximately 3ng/ml when used as a capture antibody.

靶标

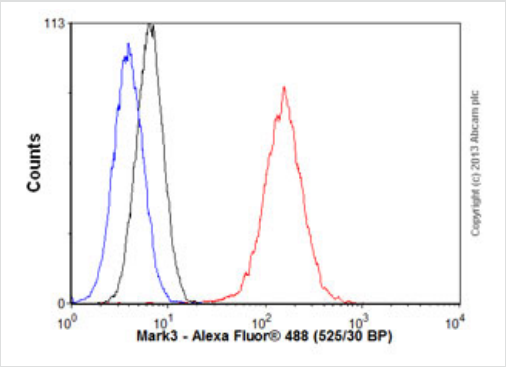
功能	Involved in the specific phosphorylation of microtubule-associated proteins for tau, MAP2 and MAP4. Phosphorylates CDC25C on 'Ser-216'.
组织特异性	Ubiquitous.
序列相似性	Belongs to the protein kinase superfamily. CAMK Ser/Thr protein kinase family. MARK subfamily. Contains 1 KA1 (kinase-associated) domain.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

图片



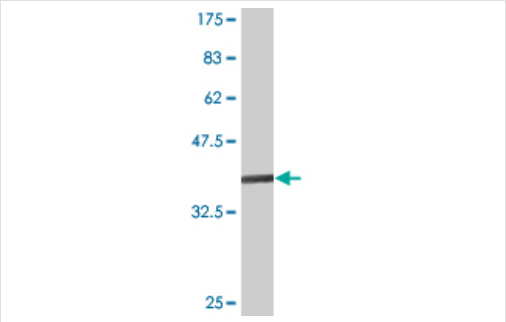
Western blot - Anti-Mark3 antibody (ab77199)

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: MARK3 knockout HAP1 cell lysate (20 µg)
Lane 3: HeLa cell lysate (20 µg)
Lane 4: K562 cell lysate (20 µg)
Lanes 1 - 4: Merged (red and green) signal. Green - ab77199 observed at 85 kDa. Red - loading control, ab181602, observed at 37 kDa.
ab77199 was shown not to specifically react with MARK3, when MARK3 knockout samples were used. Wild-type and MARK3 knockout samples were subjected to SDS-PAGE. ab77199 and ab181602 (loading control to GAPDH) were diluted at 1 µg/mL and 1/10 000 respectively and incubated overnight at 4°C. Blots were developed with goat anti-rabbit IgG (H + L) and goat anti-mouse IgG (H + L) secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Flow Cytometry - Anti-Mark3 antibody (ab77199)

Overlay histogram showing HeLa cells stained with ab77199 (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 (ab77199, 0.01µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-mouse IgG (H&L) (ab150113) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG2b [PLPV219] (ab91366, 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. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



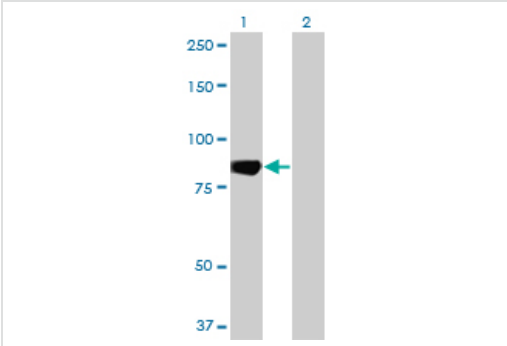
Western blot - Mark3 antibody (ab77199)

Anti-Mark3 antibody (ab77199) at 5 µg/ml + Recombinant tagged human Mark3 fragment at 0.2 µg

Secondary
Goat Anti-Mouse IgG (H&L)-HRP Conjugate at 1/5000 dilution

Predicted band size: 87 kDa
Observed band size: 37 kDa

Western blot against the tagged recombinant protein immunogen, which has a predicted molecular weight of 37 kDa.



Western blot - Mark3 antibody (ab77199)

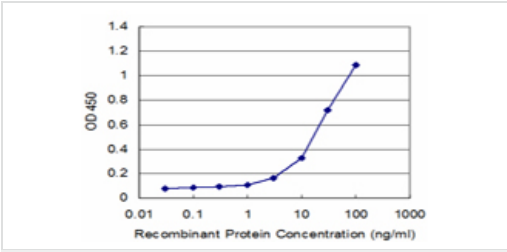
All lanes : Anti-Mark3 antibody (ab77199) at 5 µg/ml

Lane 1 : Mark3 transfected 293T cell lysate
Lane 2 : Non transfected 293T cell lysate

Lysates/proteins at 25 µg per lane.

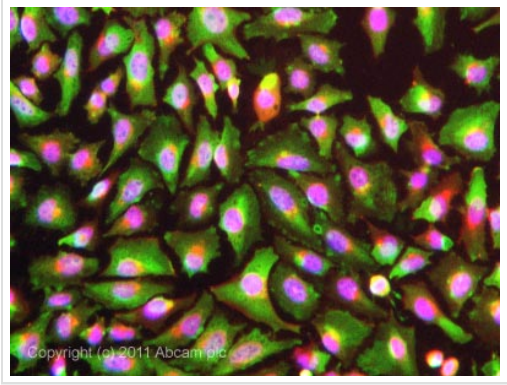
Secondary
All lanes : Goat Anti-Mouse IgG (H&L)-HRP Conjugate at 1/2500 dilution

Predicted band size: 87 kDa
Observed band size: 87 kDa



ELISA - Mark3 antibody (ab77199)

Detection limit for ab77199 is approximately 3ng/ml as a capture antibody.



Immunocytochemistry/ Immunofluorescence-Mark3 antibody(ab77199)

ICC/IF image of ab77199 stained HeLa cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab77199, 5µg/ml) overnight at +4°C. The secondary antibody (green) was ab96879, DyLight® 488 goat anti-mouse IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.

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