

## Product datasheet

# Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] ab32519

RabMAb<sup>®</sup>

★★★★☆ 2 Abreviews 5 References 8 Images

### Overview

<b>Product name</b>	Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336]
<b>Description</b>	Rabbit monoclonal [YE336] to Myosin Phosphatase 1+Myosin Phosphatase 2
<b>Host species</b>	Rabbit
<b>Specificity</b>	This antibody is specific for human Myosin Phosphatase 1 and Myosin Phosphatase 2.
<b>Tested applications</b>	<b>Suitable for:</b> WB, ICC/IF, Flow Cyt, IP
<b>Species reactivity</b>	<b>Reacts with:</b> Mouse, Rat, Human
<b>Immunogen</b>	Synthetic peptide within Human Myosin Phosphatase 1+Myosin Phosphatase 2 (C terminal). The exact sequence is proprietary.
<b>Positive control</b>	WB: HeLa, Jurkat, 293T, MCF-7, NIH/3T3 and C6 whole cell lysate. ICC/IF: HEK293 and NIH/3T3 cells FC: HEK293 and HeLa cells,
<b>General notes</b>	<p>Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <a href="#">RabMAb<sup>®</sup> patents</a>.</p> <p><b>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.</b></p>

### Properties

<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
<b>Storage buffer</b>	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.21% BSA
<b>Purity</b>	Protein A purified
<b>Clonality</b>	Monoclonal

Clone number YE336  
Isotype IgG

## Applications

Our [Abpromise guarantee](#) covers the use of **ab32519** 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
WB	★★★★☆	1/2000. Predicted molecular weight: 110 kDa. <b>For unpurified use at 1/5000.</b>
ICC/IF	★★★★☆	1/100.
Flow Cyt		1/40 - 1/100. <a href="#">ab172730</a> - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IP		1/30. <b>For unpurified use at 1/80.</b>

## Target

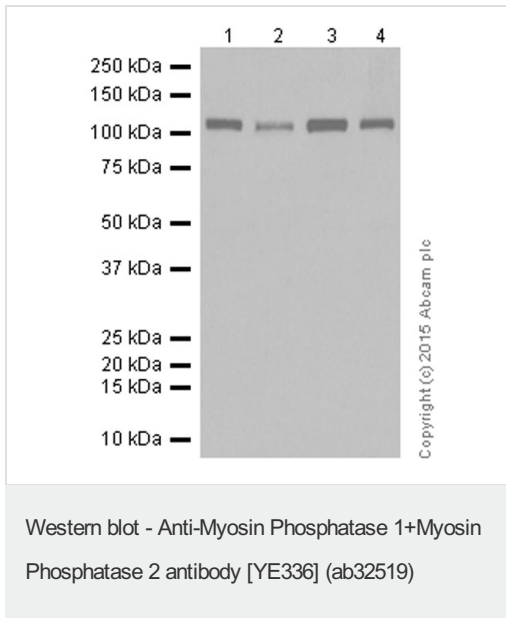
### Relevance

Myosin phosphatase regulates the interaction of actin and myosin downstream of the guanosine triphosphatase Rho. The small guanosine triphosphatase Rho is implicated in myosin light chain (MLC) phosphorylation, which results in contraction of smooth muscle and interaction of actin and myosin in nonmuscle cells. The guanosine triphosphate (GTP)-bound, active form of RhoA (GTP.RhoA) specifically interacted with the myosin-binding subunit (MBS) of myosin phosphatase, which regulates the extent of phosphorylation of MLC. Rho-associated kinase (Rho-kinase), which is activated by GTP. RhoA, phosphorylated MBS and consequently inactivated myosin phosphatase. Overexpression of RhoA or activated RhoA in NIH 3T3 cells increased phosphorylation of MBS and MLC. Thus, Rho appears to inhibit myosin phosphatase through the action of Rho-kinase.

### Cellular localization

Cytoplasmic; along actomyosin filaments and stress fibers.

## Images



**All lanes :** Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519) at 1/2000 dilution

**Lane 1 :** HeLa whole cell lysate

**Lane 2 :** Jurkat whole cell lysate

**Lane 3 :** 293T whole cell lysate

**Lane 4 :** MCF-7 whole cell lysate

Lysates/proteins at 20 µg per lane.

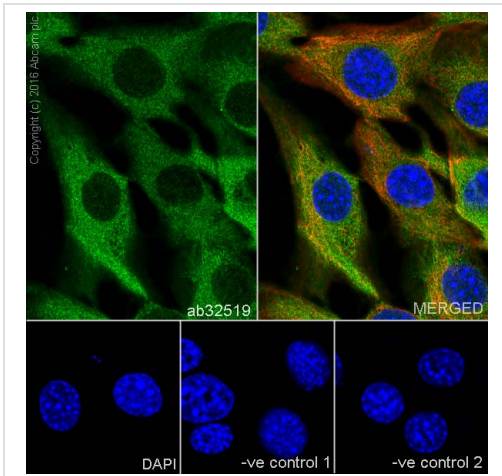
#### Secondary

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

**Predicted band size:** 110 kDa

**Observed band size:** 110 kDa

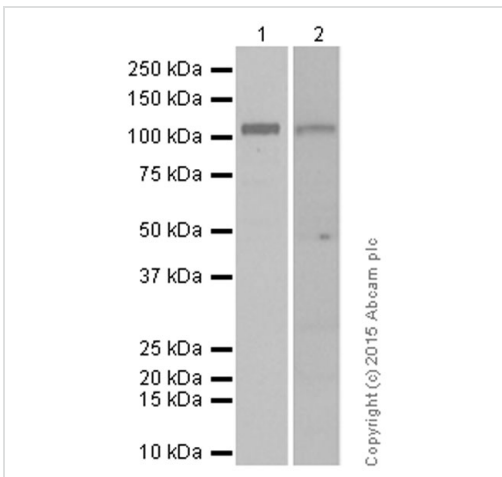
Blocking and Diluting buffer 5% NFDm/TBST



Immunocytochemistry/ Immunofluorescence - Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519)

Immunocytochemistry/Immunofluorescence analysis of NIH/3T3 cells labelling Myosin Phosphatase 1+Myosin Phosphatase 2 with purified ab32519 at 1/100. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% Triton X-100. [ab150077](#), Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. Cells were counter-stained with [ab7291](#) anti-Tubulin (mouse mAb) followed by [ab150120](#), AlexaFluor®594 goat anti-mouse secondary both at 1/1000. Nuclei were counterstained with DAPI (blue).

For negative control 1, rabbit primary antibody was used followed by anti-mouse secondary antibody ([ab150120](#)). For negative control 2, [ab7291](#) (mouse primary antibody) was used followed by anti-rabbit secondary antibody ([ab150077](#)).



Western blot - Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519)

**All lanes :** Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519) at 1/2000 dilution

**Lane 1 :** NIH/3T3 whole cell lysate

**Lane 2 :** C6 whole cell lysate

Lysates/proteins at 20 µg per lane.

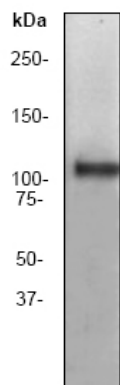
**Secondary**

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

**Predicted band size:** 110 kDa

**Observed band size:** 110 kDa

Blocking and Diluting buffer 5% NFDN/TBST

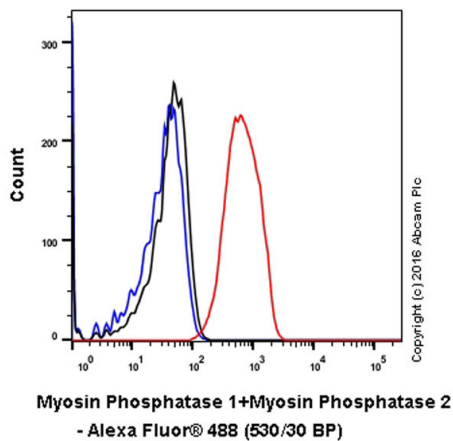


Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519) at 1/5000 dilution (unpurified) + 293T cell lysate.

**Predicted band size:** 110 kDa

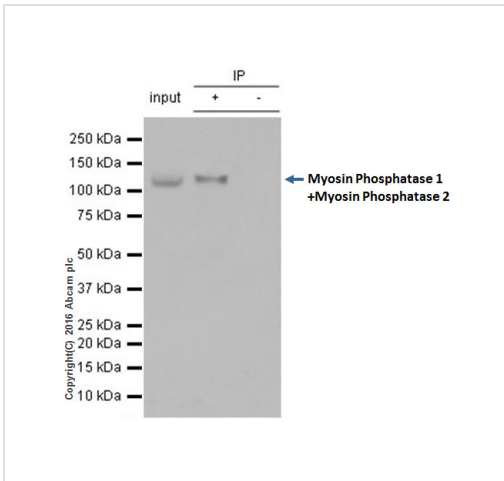
**Observed band size:** 110 kDa

Western blot - Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519)



Flow cytometry analysis of HeLa cells labelling Myosin Phosphatase 1+Myosin Phosphatase 2 (red) with purified ab32519 at dilution of 1/40. The secondary antibody used was goat anti rabbit IgG (FITC) at 1/500. Cells were fixed with 4% paraformaldehyde. Isotype control antibody was Rabbit monoclonal IgG (black). The blue line shows cells without incubation with primary antibody and secondary antibody.

Flow Cytometry - Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519)



Immunoprecipitation - Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519)

ab32519 at 1/30 dilution immunoprecipitating Myosin Phosphatase 1+Myosin Phosphatase 2 in HeLa whole cell lysate observed at 110 kDa (lanes 1 and 2).

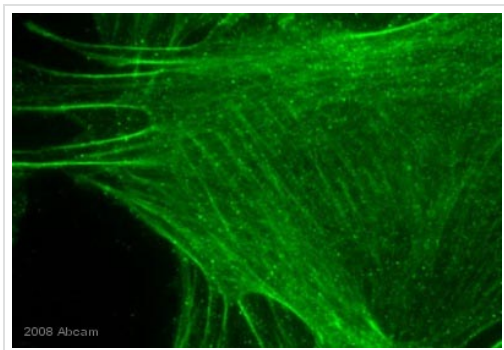
Lane 1 (input): HeLa whole cell lysate 10ug

Lane 2 (+): ab32519 + HeLa whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab32519 in HeLa whole cell lysate

For western blotting, ab32519 was used followed by VeriBlot for IP Detection Reagent (HRP) (ab131366) for detection at a dilution of 1/10,000.

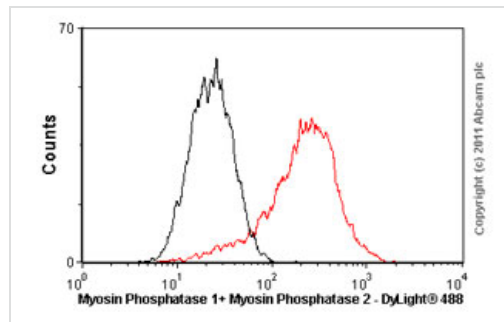
Blocking and Diluting buffer and concentration: 5% NFD/MTBST.



Immunocytochemistry/ Immunofluorescence - Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519)

This image is courtesy of an anonymous Abreview

ab32519 staining mouse fibroblast cells by ICC/IF. Cells were PFA fixed, permeabilized in Triton X-100 and blocked in 1% BSA for 30 minutes at 25°C. The primary antibody was diluted 1/200 and incubated with sample for 4 hours at 25°C. An Alexa Fluor® 488 conjugated goat monoclonal to rabbit, diluted 1/500 was used as the secondary.



Flow Cytometry - Anti-Myosin Phosphatase 1+Myosin Phosphatase 2 antibody [YE336] (ab32519)

Overlay histogram showing HEK293 cells stained with unpurified ab32519 (red line). The cells were fixed with 4% paraformaldehyde (10 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 (ab32519, 1/100 dilution) 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. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HEK293 cells fixed with 100% methanol (5 min)/permeabilized in 0.1% PBS-Tween used under the same conditions.

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