Product datasheet

Anti-Calponin 1 antibody [EP798Y] ab46794

Overview

Product name: Anti-Calponin 1 antibody [EP798Y]
Description: Rabbit monoclonal [EP798Y] to Calponin 1
Host species: Rabbit
Tested applications: Suitable for: WB, IHC-P, ICC/IF
Unsuitable for: Flow Cyt
Species reactivity: Reacts with: Mouse, Rat, Human, Pig
Predicted to work with: Sheep
Immunogen: Synthetic peptide within Human Calponin 1 aa 250-350 (C terminal). The exact sequence is proprietary.
Database link: P51911
Positive control: ICC/IF: C2C12 and HeLa cells; Pig and mouse aortic smooth muscle cells. IHC-P: Rat lung tissue; Mouse cardiac muscle tissue; Human lung carcinoma, kidney, lung, tonsil, uterus, smooth muscle and skeletal muscle tissues. WB: Human bladder lysates, Pig heart lysates, Mouse bladder lysates and Rat bladder lysates.
General notes: 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.

This product is a recombinant rabbit monoclonal antibody.

Properties

Form: Liquid
Dissociation constant (K_D): K_D = 1.73 x 10^-10 M

20 Abreviews  133 References  20 Images
Storage buffer
pH: 7.20  
Preservative: 0.01% Sodium azide  
Constituents: PBS, 40% Glycerol, 0.05% BSA

Purity  
Protein A purified

Clonality  
Monoclonal

Clone number  
EP798Y

Isotype  
IgG

Applications
Our Abpromise guarantee covers the use of ab46794 in the following tested applications.  
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>WB</td>
<td>⭐⭐⭐⭐⭐</td>
<td>1/5000. Detects a band of approximately 34 kDa. For unpurified use at 1/20,000.</td>
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<tr>
<td>IHC-P</td>
<td>⭐⭐⭐⭐⭐</td>
<td>1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. For unpurified use at 1/100 - 1/250.</td>
</tr>
<tr>
<td>ICC/IF</td>
<td>⭐⭐⭐⭐⭐</td>
<td>1/500. For unpurified use at 1/100 - 1/250.</td>
</tr>
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</table>

Application notes  
Is unsuitable for Flow Cyt.

Target

Function  
Thin filament-associated protein that is implicated in the regulation and modulation of smooth muscle contraction. It is capable of binding to actin, calmodulin, troponin C and tropomyosin. The interaction of calponin with actin inhibits the actomyosin Mg-ATPase activity.

Tissue specificity  
Smooth muscle, and tissues containing significant amounts of smooth muscle.

Sequence similarities  
Belongs to the calponin family.  
Contains 3 calponin-like repeats.  
Contains 1 CH (calponin-homology) domain.

Images
Immunocytochemistry/Immunofluorescence analysis of C2C12 (Mouse myoblasts myoblast) cells labeling Calponin 1 with purified ab46794 at 1/500 dilution. Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Cells were counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 μg/ml). ab150077 Goat anti rabbit IgG(Alexa Fluor® 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

All lanes: Anti-Calponin 1 antibody [EP798Y] (ab46794) at 1/50000 dilution (purified)

Lane 1: Human bladder lysates
Lane 2: Mouse bladder lysates
Lane 3: Rat bladder lysates

Lysates/proteins at 15 μg per lane.

Secondary

All lanes: Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Blocking and diluting buffer: 5% NFDM/TBST.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human lung carcinoma tissue sections labeling Calponin 1 with purified ab46794 at a 1:1000 dilution. Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used.

PBS instead of the primary antibody was used as the negative control (inset).

**All lanes**: Anti-Calponin 1 antibody [EP798Y] (ab46794)

**Lanes 1-2**: Non-Failing pig LV-myocardium tissue lysate.

**Lanes 3-4**: Failing pig LV-myocardium tissue lysate injected with DOX.

Ab46794 was used to stain Calponin 1 in Neonatal piglets injected with Dox (Failing LV myocardium) and PBS (Non-Failing LV myocardium). At the protein level a more than twofold increase in Calponin 1 was observed in Dox-injected animals compared to controls (PBS).
Paraformadehyde-fixed, 0.25% Triton X-100 permeabilized mouse thoracic aortic smooth muscle cells labeling Calponin 1 using ab46794 at 1/100 dilution in ICC/IF, followed by a Goat Anti-Rabbit IgG H&L (Alexa Fluor 488) (ab150077) at 1/400 dilution.

1.5% BSA used used as blocking agent for 30 minutes at 25°C. Incubated with primary antibody for 24 hours at 4°C.

VSMCs were seeded to 35-mm plates in a low density avoiding overlapping of cells. After fixation, VSMCs were treated with 0.25% Triton X-100 for 20 minutes.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse cardiac muscle tissue sections labeling Calponin 1 with purified ab46794 at 1:1000 dilution. Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with hematoxylin.

ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used.

PBS instead of the primary antibody was used as the negative control (inset).

Anti-Calponin 1 antibody [EP798Y] (ab46794) at 1/5000 dilution (purified) + Pig heart lysates at 15 µg

**Secondary**

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

Blocking and diluting buffer: 5% NFDM/TBST.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat lung tissue sections labeling Calponin 1 with purified ab46794 at 1:1000 dilution. Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody. PBS instead of the primary antibody was used as the negative control (inset).

Unpurified ab46794 staining Calponin 1 in porcine aortic smooth muscle cells by Immunocytochemistry/Immunofluorescence. The cells were paraformaldehyde fixed, permeabilized in 0.1% Triton X-100. Samples were then incubated with primary antibody at 1/50 for 1 hour at 25°C. The secondary antibody used was ab6717 Goat polyclonal to Rabbit IgG - H&L (FITC) (green) used at a 1/400 dilution.

Representative photomicrograph of UT-myo cells (Left panel) and uterine myometrium (Right panel) stained with smooth muscle cell markers, alpha-SMA (red) and ab46794 (green) and DAPI (blue). UT-myo cells and whole-mount uterine tissue were collected from day 19 of mouse pregnancy. The placenta and embryo were removed from whole-mount tissue sections.

For full details please see paper.
Other - Anti-Calponin 1 antibody [EP798Y] (ab46794)

Anti-Calponin 1 antibody [EP798Y] (ab46794) at 1/20000 dilution (unpurified) + Human bladder lysate at 10 µg

Secondary
Goat anti-rabbit HRP at 1/2000 dilution

Observed band size: 34 kDa

why is the actual band size different from the predicted?

Immunocytochemistry/ Immunofluorescence - Anti-Calponin 1 antibody [EP798Y] (ab46794)

ICC/IF image of unpurified ab46794 stained HeLa (Human epithelial cell line from cervix adenocarcinoma) cells.

Cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilize the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab46794, 5µg/ml) overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti-rabbit IgG - H&L, pre-adsorbed (ab96899) 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.
Immunohistochemical staining of paraffin-embedded human smooth muscle using unpurified ab46794 at 1/100 dilution

All lanes: Anti-Calponin 1 antibody [EP798Y] (ab46794) at 1/20000 dilution (unpurified)

All lanes: Whole tissue lysate prepared from bovine lymph vessels

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP conjugated goat anti-rabbit polyclonal at 1/5000 dilution

Developed using the ECL technique.

**Observed band size:** 34 kDa. Why is the actual band size different from the predicted?

**Exposure time:** 1 minute
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calponin 1 antibody [EP798Y] (ab46794)

Unpurified ab46794 showing positive staining in normal lung vessel tissue.

Unpurified ab46794 showing positive staining in normal kidney vessels tissue.

Unpurified ab46794 showing positive staining in normal tonsil vessel tissue.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Calponin 1 antibody [EP798Y] (ab46794)

Unpurified ab46794 showing positive staining in normal uterus tissue.

Unpurified ab46794 showing negative staining in skeletal muscle tissue.

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