# abcam

# Product datasheet

# Anti-Actin antibody [EPR16769] - BSA and Azide free ab219733



# 6 References 9 Images

#### Overview

Product name Anti-Actin antibody [EPR16769] - BSA and Azide free

**Description** Rabbit monoclonal [EPR16769] to Actin - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, ICC/IF, IP, Flow Cyt (Intra)

Species reactivity Reacts with: Mouse, Rat, Chicken, Human

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HeLa, 293T, C6, RAW 264.7, PC-12, NIH/3T3 and UMNSAH/DF-1 whole cell lysates;

Human skeletal muscle, fetal spleen, fetal brain, fetal heart, fetal kidney and cardiac muscle lysates; Mouse and Rat brain, heart, kidney and spleen lysates. IHC-P: Human prostate hyperplasia, Mouse skeletal muscle and Rat skeletal muscle tissues. ICC/IF: NIH/3T3 cells. IP:

NIH/3T3 whole cell extract. Flow Cyt (intra): HeLa cells

**General notes** ab219733 is the carrier-free version of **ab179467**.

Our <u>carrier-free</u> 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 <u>conjugation kits</u> 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<sup>®</sup> Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> 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.

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Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

ClonalityMonoclonalClone numberEPR16769

**Isotype** IgG

# **Applications**

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab219733 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		Use at an assay dependent concentration. Detects a band of approximately 42 kDa (predicted molecular weight: 42 kDa).
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.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration.

# Target

**Function** Actins are highly conserved proteins that are involved in various types of cell motility and are

ubiquitously expressed in all eukaryotic cells.

Involvement in disease Defects in ACTA1 are the cause of nemaline myopathy type 3 (NEM3) [MIM:161800]. A form of

nemaline myopathy. Nemaline myopathies are muscular disorders characterized by muscle weakness of varying severity and onset, and abnormal thread-or rod-like structures in muscle fibers on histologic examination. The phenotype at histological level is variable. Some patients present areas devoid of oxidative activity containg (cores) within myofibers. Core lesions are

unstructured and poorly circumscribed.

Defects in ACTA1 are a cause of myopathy congenital with excess of thin myofilaments

(MPCETM) [MIM:161800]. A congenital muscular disorder characterized at histological level by areas of sarcoplasm devoid of normal myofibrils and mitochondria, and replaced with dense masses of thin filaments. Central cores, rods, ragged red fibers, and necrosis are absent. Defects in ACTA1 are a cause of congenital myopathy with fiber-type disproportion (CFTD) [MIM:255310]; also known as congenital fiber-type disproportion myopathy (CFTDM). CFTD is a genetically heterogeneous disorder in which there is relative hypotrophy of type 1 muscle fibers compared to type 2 fibers on skeletal muscle biopsy. However, these findings are not specific and can be found in many different myopathic and neuropathic conditions.

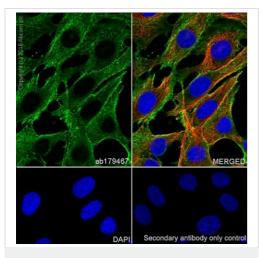
Sequence similarities

**Cellular localization** 

Belongs to the actin family.

Cytoplasm > cytoskeleton.

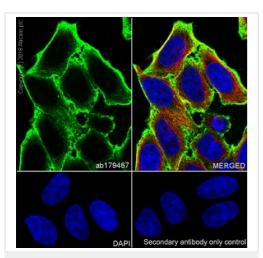
#### **Images**



Immunocytochemistry/ Immunofluorescence - Anti-Actin antibody [EPR16769] - BSA and Azide free (ab219733)

Immunocytochemistry/ Immunofluorescence analysis of NIH/3T3 (Mouse embryonic fibroblast) cells labeling Actin with Purified  $\underline{ab179467}$  at 1:100 dilution (6.98  $\mu g/ml$ ). 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  $\mu g/ml$ ). Goat anti rabbit lgG (Alexa Fluor  $^{\&}$  488,  $\underline{ab150077}$ ) was used as the secondary antibody at 1:1000 dilution (2  $\mu g/ml$ ) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

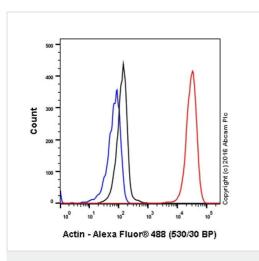
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab179467</u>).



Immunocytochemistry/ Immunofluorescence - Anti-Actin antibody [EPR16769] - BSA and Azide free (ab219733)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelia cell) cells labeling Actin with Purified  $\underline{ab179467}$  at 1:100 dilution ( 6.98  $\mu$ g/ml). 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  $\mu$ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488,  $\underline{ab150077}$ ) was used as the secondary antibody at 1:1000 dilution (2  $\mu$ g/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

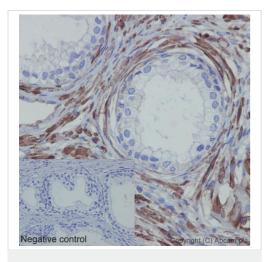
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab179467</u>).



Flow Cytometry (Intracellular) - Anti-Actin antibody [EPR16769] - BSA and Azide free (ab219733)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labelling. Actin with purified <u>ab179467</u> at 1/70 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. An Alexa Fluor<sup>®</sup>488-conjugated goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. 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 (<u>ab179467</u>).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Actin antibody

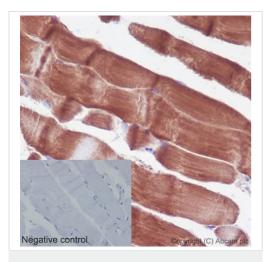
[EPR16769] - BSA and Azide free (ab219733)

Immunohistochemical analysis of paraffin-embedded Human prostate hyperplasia tissue labeling Actin with <u>ab179467</u> at 1/1000 dilution, followed by prediluted HRP Polymer for Rabbit/Mouse IgG. Cytoplasm staining on smooth muscle cells is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary ab, secondary ab is prediluted HRP Polymer for Rabbit/Mouse IgG.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab179467</u>).

Heat mediated antigen retrieval was performed with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Actin antibody

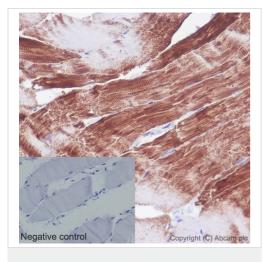
[EPR16769] - BSA and Azide free (ab219733)

Immunohistochemical analysis of paraffin-embedded Mouse skeletal muscle tissue labeling Actin with <u>ab179467</u> at 1/1000 dilution, followed by prediluted HRP Polymer for Rabbit/Mouse IgG. Cytoplasm staining is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary ab, secondary ab is prediluted HRP Polymer for Rabbit/Mouse IgG.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab179467</u>).

Heat mediated antigen retrieval was performed with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Actin antibody

[EPR16769] - BSA and Azide free (ab219733)

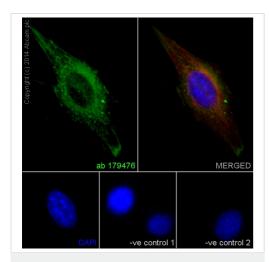
Immunohistochemical analysis of paraffin-embedded Rat skeletal muscle tissue labeling Actin with <u>ab179467</u> at 1/1000 dilution, followed by prediluted HRP Polymer for Rabbit/Mouse IgG.

Cytoplasm staining is observed. Counter stained with Hematoxylin.

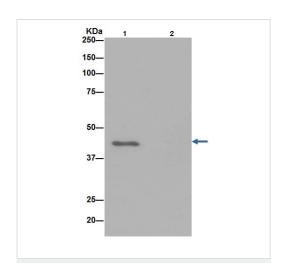
Negative control: Using PBS instead of primary ab, secondary ab is prediluted HRP Polymer for Rabbit/Mouse IgG.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab179467).

Heat mediated antigen retrieval was performed with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Actin antibody [EPR16769] - BSA and Azide free (ab219733)



Immunoprecipitation - Anti-Actin antibody

[EPR16769] - BSA and Azide free (ab219733)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embyro fibroblast cells) cells labeling Actin with <a href="mailto:ab179467">ab179467</a> at 1/50 dilution, followed by Goat <a href="mailto:anti-rabbit Alexa Fluor® 488">anti-rabbit Alexa Fluor® 488</a> (lgG) (<a href="mailto:ab150077">ab150077</a>) secondary antibody at 1/200 dilution (green). Cytoplasm staining on NIH/3T3 cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with <a href="mailto:ab7291">ab7291</a> (anti-Tubulin mouse mAb) at 1/500 dilution and <a href="mailto:ab150120">ab150120</a> (goat <a href="mailto:anti-mouse AlexaFluor®594">anti-mouse AlexaFluor®594</a> secondary) at 1/500 dilution (red).

The negative controls are as follows;

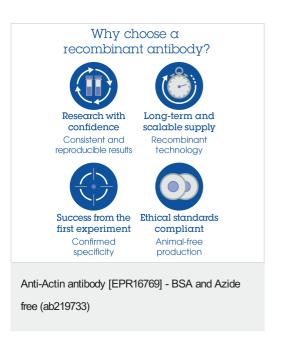
- 1. <u>ab179467</u> at 1/50 dilution followed by <u>ab150120</u> (goat <u>antimouse AlexaFluor®594</u> secondary) at 1/500 dilution.
- 2. <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/500 dilution followed by <u>ab150077</u> (goat <u>anti-rabbit Alexa Fluor®488</u> (lgG H&L) at 1/200 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab179467</u>).

Actin was immunoprecipitated from 1mg of NIH/3T3 (Mouse embyro fibroblast cells) whole cell extract with <u>ab179467</u> at 1/40 dilution. Western blot was performed from the immunoprecipitate using <u>ab179467</u> at 1/1000 dilution. Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated was used as secondary antibody at 1/1000 dilution. Lane 1: NIH/3T3 whole cell extract. Lane 2: PBS instead of NIH/3T3 whole cell extract.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab179467).



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