# abcam

**Product datasheet** 

# Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free ab242395

КО 評価済 มาวนราวง RabMAb

★★★★★ <u>2 Abreviews</u> 画像数 13

Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free
Rabbit monoclonal [SP171] to alpha smooth muscle Actin - BSA and Azide free
Rabbit
適用あり: ICC/IF, IHC-P, WB, Flow Cyt (Intra), mIHC
交差種: Mouse, Rat, Human
交差が予測される動物種: Rabbit, Chicken, Cow, Pig 🛛 🐴
Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
IHC-P: Human colon, Mouse colon, and Rat colon tissue. WB: Recombinant Human alpha smooth muscle Actin protein ( <b>ab114148</b> ); HeLa cell lysate. Flow Cyt (intra): HeLa, NIH/3T3 and C6 cells. ICC: NIH/3T3 and HeLa cells.
ab242395 is the carrier-free version of <u>ab150301</u> .
Our <b><u>carrier-free</u></b> 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><b>conjugation kits</b></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 <u>see here</u> .

### 製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C. Do Not Freeze.
バッファー	pH: 7.20 Constituent: PBS
キャリア・フリー	はい
精製度	Protein A purified
特記事項(精製)	Purified from TCS by protein A/G.
ポリ/モノ	モノクローナル
クローン名	SP171
アイソタイプ	lgG

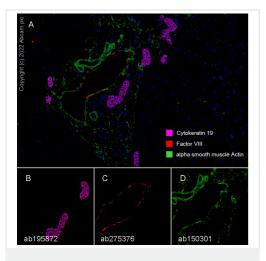
アプリケーション

The Abpromise guaranteeAbpromise保証は、<br/>次のテスト済みアプリケーションにおけるab242395の使用に適用されますアプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
ICC/IF		Use at an assay dependent concentration.
IHC-P	★ ★ ★ ★ ★ <u>(2)</u>	Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 42 kDa.
Flow Cyt (Intra)		Use at an assay dependent concentration.
mIHC		Use at an assay dependent concentration.

ターゲット情報	
機能	Actins are highly conserved proteins that are involved in various types of cell motility and are ubiquitously expressed in all eukaryotic cells.
関連疾患	Defects in ACTA2 are the cause of aortic aneurysm familial thoracic type 6 (AAT6) [MIM:611788]. AATs are characterized by permanent dilation of the thoracic aorta usually due to degenerative changes in the aortic wall. They are primarily associated with a characteristic histologic appearance known as 'medial necrosis' or 'Erdheim cystic medial necrosis' in which there is degeneration and fragmentation of elastic fibers, loss of smooth muscle cells, and an accumulation of basophilic ground substance.
配列類似性	Belongs to the actin family.

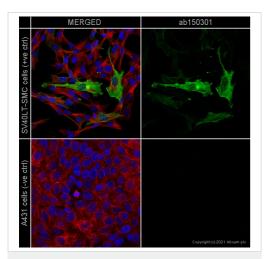
#### 画像



Multiplex immunohistochemistry - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395)

Fluorescence multiplex immunohistochemical analysis of human liver tissue (formalin-fixed paraffin-embedded section). Panel A shows merged staining of ab195872 anti-Cytokeratin 19 stained on branch of bile ducts (magenta; Opal™690) at 1:8000 ( 0.127 µg/ml) [Panel B], ab275376 anti-Factor VIII stained on endothelial cells (red; Opal<sup>™</sup>570) at 1:1000 ( 0.457 µg/ml) [Panel C], and **ab150301** anti-alpha smooth muscle Actin stained on smooth muscles (green; Opal<sup>™</sup>520) at 1:200 (0.14 µg/ml) [Panel C] on human liver. DAPI was used as a nuclear counter stain. Followed by Opal Polymer HRP Ms + Rb secondary. The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal<sup>™</sup> 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope. The section was incubated in three rounds of staining: in the order of ab195872 for 30 mins, ab275376 for 30 mins and ab150301 for 10 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins was used.

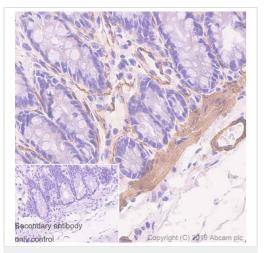
This data was developed using <u>ab150301</u>, the same antibody clone in a different buffer formulation.



Immunocytochemistry/ Immunofluorescence - Antialpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395)

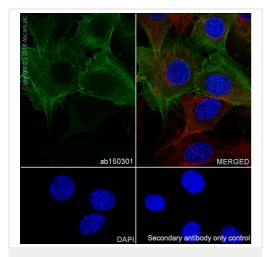
This data was developed using the same antibody clone in a different buffer formulation (**ab150301**). **ab150301** staining alpha smooth muscle Actin in SV40LT-SMC cells (positive control, top panel) and A431 cells (negative control, bottom panel). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab150301** at 1µg/ml concentration and **ab7291** (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor<sup>®</sup> 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor<sup>®</sup> 594) (**ab150120**) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).

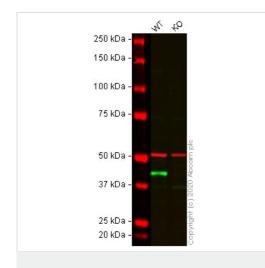


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat colon tissue sections labeling alpha smooth muscle Actin with **ab150301** at 1/200 dilution (0.26 µg/ml). Heat mediated antigen retrieval was performed Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 10 mins. Goat Anti-Rabbit & Mouse IgG (HRP) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab150301**)



Immunocytochemistry/ Immunofluorescence - Antialpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395) Immunocytochemistry/ Immunofluorescence analysis of NIH/3T3 (mouse embryonic fibroblast) cells labeling alpha smooth muscle Actin with purified <u>ab150301</u> at 1/100(1.65  $\mu$ g/ml). Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Cells were counterstained with <u>ab195889</u> Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) 1/200 (2.5  $\mu$ g/ml). Goat anti rabbit IgG (Alexa Fluor<sup>®</sup> 488, <u>ab150077</u>) was used as the secondary antibody at 1/1000 (2  $\mu$ g/ml) dilution. DAPI (blue) was used as the secondary antibody 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>ab150301</u>).



Western blot - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395) All lanes : Anti-alpha smooth muscle Actin antibody [SP171] (ab150301) at 1/130 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate Lane 2 : ACTA2 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lysates/proteins at 20 µg per lane.

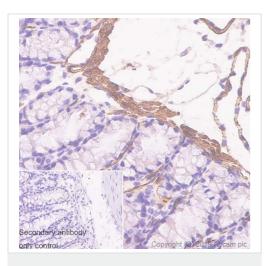
Performed under reducing conditions.

Predicted band size: 42 kDa Observed band size: 42 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab150301**).

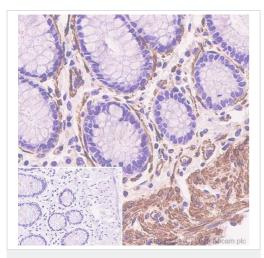
Lanes 1 - 2: Merged signal (red and green). Green - <u>ab150301</u> observed at 42 kDa. Red - loading control, <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

**ab150301** was shown to react with alpha smooth muscle Actin in wild-type HeLa cells in western blot. Loss of signal was observed when ACTA2 knockout sample was used. Wild-type HeLa and ACTA2 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween<sup>®</sup>) before incubation with **ab150301** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4°C at a 1 in 130 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



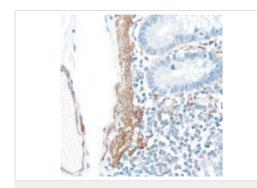
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse colon tissue sections labeling alpha smooth muscle Actin with <u>ab150301</u> at 1/200 dilution (0.26 µg/ml). Heat mediated antigen retrieval was performed Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 10 mins. Goat Anti-Rabbit & Mouse IgG (HRP) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150301</u>)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395)

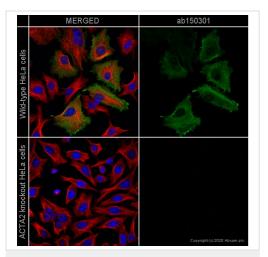
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human colon tissue sections labeling alpha smooth muscle Actin with <u>ab150301</u> at 1/200 dilution (0.26 µg/ml). Heat mediated antigen retrieval was performed Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 10 mins. Goat Anti-Rabbit & Mouse IgG (HRP) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150301</u>)



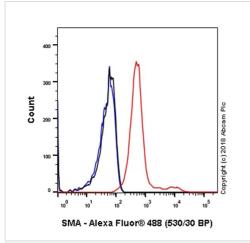
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395)

Immunohistochemical analysis of formalin fixed, paraffin embedded human colon tissue labeling alpha smooth muscle Actin with <u>ab150301</u> at 1/200 dilution.

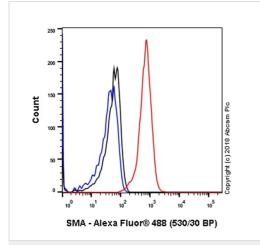
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA and sodium azide (<u>ab150301</u>)



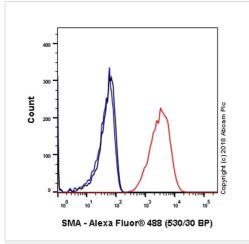
Immunocytochemistry/ Immunofluorescence - Antialpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395) This data was developed using the same antibody clone in a different buffer formulation (ab150301). ab150301 staining alpha smooth muscle Actin in wild-type HeLa cells (top panel) and ACTA2 knockout HeLa cells (bottom panel). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab150301 at 5µg/ml concentration and ab7291 (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor<sup>®</sup> 488) (ab150081) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor<sup>®</sup> 594) (ab150120) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI. Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).



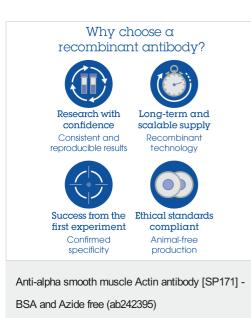
Flow Cytometry (Intracellular) - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395) Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma epithelial cell) cells labeling alpha smooth muscle Actin with purified <u>ab150301</u> at 1/200 dilution (0.825µg/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 488, <u>ab150077</u>) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal lgG (<u>ab172730</u>) / Black. Unlabeled control - Unlabelled cells / blue. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab150301</u>).



Flow Cytometry (Intracellular) - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395) Intracellular Flow Cytometry analysis of C6 (Rat glial tumor glial cell) cells labeling alpha smooth muscle Actin with purified **ab150301** at 1/200 dilution (0.825µg/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor<sup>®</sup> 488, **ab150077**) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal IgG (**ab172730**) / Black. Unlabeled control - Unlabelled cells / blue. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab150301**).



Flow Cytometry (Intracellular) - Anti-alpha smooth muscle Actin antibody [SP171] - BSA and Azide free (ab242395)



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Intracellular Flow Cytometry analysis of NIH/3T3 (Mouse embryonic fibroblast) cells labeling alpha smooth muscle Actin with purified **ab150301** at 1/200 dilution (0.825µg/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 488, **ab150077**) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal lgG (**ab172730**) / Black. Unlabeled control - Unlabelled cells / blue. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab150301**).

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