abcam

Product datasheet

Anti-Caveolin-1 antibody [EPR15554] - BSA and Azide free ab240332





RabMAb

11 图像

概述

产品名称 Anti-Caveolin-1抗体[EPR15554] - BSA and Azide free

描述 兔单克隆抗体[EPR15554] to Caveolin-1 - BSA and Azide free

宿主 Rabbit

经测试应用 适用于: ICC/IF, Flow Cyt (Intra), IP, IHC-P, WB

种属反应性 与反应: Mouse, Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: A549, A431 and HeLa cell lysates. ICC/IF: HeLa and A763 cells. IHC-P: Human liver and

squamous cell carcinoma of cervix tissue; Mouse lung tissue. Flow Cyt (intra): NIH3T3 and HeLa

cells.

常规说明 ab240332 is the carrier-free version of ab192869.

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 **conjugation kits** 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[®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] 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.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit

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性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C. Do Not Freeze.

存储溶液 pH: 7.2

Constituent: PBS

无载体 **是**

纯**度** Protein A purified

克隆 单克隆

克隆编号 EPR15554

同种型 IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab240332于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration. <u>ab199376</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
IP		Use at an assay dependent concentration.
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.
WB		Use at an assay dependent concentration. Detects a band of approximately 17, 20 kDa (predicted molecular weight: 17, 20 kDa).

靶你

功能 May act as a scaffolding protein within caveolar membranes. Interacts directly with G-protein

alpha subunits and can functionally regulate their activity (By similarity). Involved in the

costimulatory signal essential for T-cell receptor (TCR)-mediated T-cell activation. Its binding to DPP4 induces T-cell proliferation and NF-kappa-B activation in a T-cell receptor/CD3-dependent

manner. Recruits CTNNB1 to caveolar membranes and may regulate CTNNB1-mediated

signaling through the Wnt pathway.

组织特异性 Expressed in muscle and lung, less so in liver, brain and kidney.

疾病相关 Defects in CAV1 are the cause of congenital generalized lipodystrophy type 3 (CGL3)

[MIM:612526]; also called Berardinelli-Seip congenital lipodystrophy type 3 (BSCL3). Congenital generalized lipodystrophies are autosomal recessive disorders characterized by a near absence of adipose tissue, extreme insulin resistance, hypertriglyceridemia, hepatic steatosis and early

onset of diabetes.

序列相似性 Belongs to the caveolin family.

翻译后修饰 The initiator methionine for isoform Beta is removed during or just after translation. The new N-

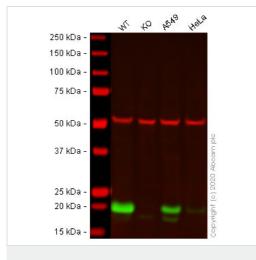
terminal amino acid is then N-acetylated.

细胞定位 Golgi apparatus membrane. Cell membrane. Membrane > caveola. Membrane raft. Colocalized

with DPP4 in membrane rafts. Potential hairpin-like structure in the membrane. Membrane protein

of caveolae.

图片



Western blot - Anti-Caveolin-1 antibody [EPR15554]

- BSA and Azide free (ab240332)

All lanes : Anti-Caveolin-1 antibody [EPR15554] - N-terminal (ab192869) at 1/1000 dilution

Lane 1: Wild-type A-431 (Human epidermoid carcinoma cell line)

whole cell lysate

Lane 2: CAV1 knockout A-431 (Human epidermoid carcinoma

cell line) whole cell lysate

Lane 3: A549 (Human lung carcinoma cell line) whole cell lysate

Lane 4: 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: 17, 20 kDa

Observed band size: 21-24 kDa

This data was developed using the same antibody clone in a different buffer formulation (<u>ab192869</u>).

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab192869</u> observed at 21-24 kDa. Red - loading control, <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

<u>ab192869</u> was shown to react with Caveolin-1 in A431 wild-type cells in western blot. Loss of signal was observed when CAV1 knockout sample was used. A431 wild-type and CAV1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 5% milk in TBS-T (0.1% Tween[®]) before incubation with

ab192869 and ab7291 (Mouse anti-Alpha Tubulin [DM1A] overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

MERGED ab192869

Wild-trype HeLa cells

CAVI knockout HeLa cells

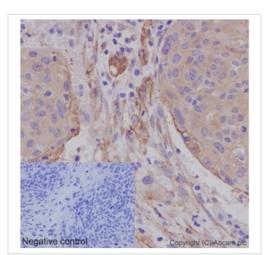
Cavilidation of the cells of t

Immunocytochemistry/ Immunofluorescence - Anti-Caveolin-1 antibody [EPR15554] - BSA and Azide free (ab240332)

This data was developed using the same antibody clone in a different buffer formulation (ab192869).

ab192869 staining Caveolin-1 in wild-type HeLa cells (top panel) and CAV1 knockout HeLa cells (ab255371) (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 ab192869 at 1/500 dilution 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 lgG (Alexa Fluor[®] 488) (ab150081) at 2 μg/ml (shown in green) and a goat secondary antibody to mouse lgG (Alexa Fluor[®] 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).



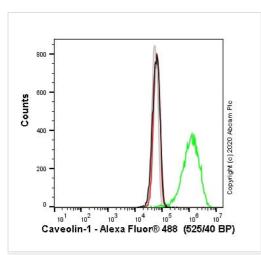
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Caveolin-1 antibody

[EPR15554] - BSA and Azide free (ab240332)

Immunohistochemical analysis of paraffin-embedded Human squamous cell carcinoma of cervix tissue labeling Caveolin-1 using **ab192869** at 1/4000 dilution (0.15 µg/ml). A prediluted HRP Polymer for Rabbit IgG was used as the secondary. Counterstain: Hematoxylin. Negative control also shown.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab192869).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



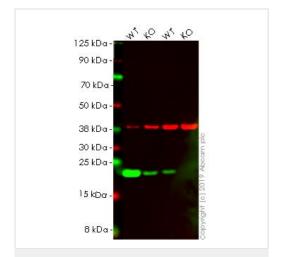
Flow Cytometry (Intracellular) - Anti-Caveolin-1 antibody [EPR15554] - BSA and Azide free (ab240332)

Intracellular Intracellular Flow Cytometry overlay histogram showing wild-type HeLa (green line) and CAV1 knockout HeLa cells (ab255371) stained with ab240332 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab240332) (1x10 6 in 100µl at 0.04 µg/ml) for 30 min at 22°C.

The secondary antibody Goat anti-rabbit lgG H&L (Alexa Fluor[®] 488, pre-adsorbed) (<u>ab150081</u>) was used at 1/2000 for 30 min at 22°C.

Isotype control antibody was Rabbit IgG (monoclonal) (<u>ab172730</u>) used at the same concentration and conditions as the primary antibody (wild-type HeLa - black line; CAV1 knockout HeLa - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.



Western blot - Anti-Caveolin-1 antibody [EPR15554]
- BSA and Azide free (ab240332)

All lanes : Anti-Caveolin-1 antibody [EPR15554] - N-terminal (ab192869) at 1/10000 dilution

Lane 1 : A431 cell lysate Lane 2 : A549 cell lysate

Lane 3: Wild-type HeLa cell lysate

Lane 4: Caveolin-1 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

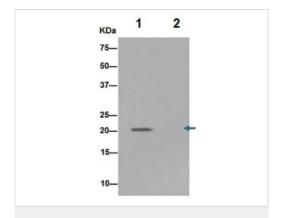
Predicted band size: 17, 20 kDa **Observed band size:** 20 kDa

This data was developed using the same antibody clone in a different buffer formulation (<u>ab192869</u>).

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab192869</u> observed at 20 kDa. Red - loading control, <u>ab8245</u> observed at 37

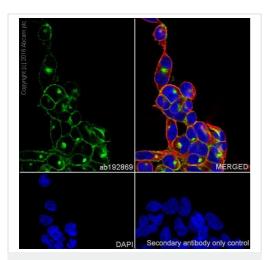
kDa.

<u>ab192869</u> was shown to react with Caveolin-1 in wild-type HeLa. Loss of signal was observed when knockout cell line <u>ab255371</u> (knockout cell lysate <u>ab263806</u>) was used. Wild-type and Caveolin-1 knockout samples were subjected to SDS-PAGE. <u>ab192869</u> and Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) were incubated overnight at 4°C at 1 in 10000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunoprecipitation - Anti-Caveolin-1 antibody [EPR15554] - BSA and Azide free (ab240332) Immunoprecipitation analysis of A431 cell lysate labeling Caveolin-1 using **ab192869** at 1/30 dilution (Lane 1). PBS negative control (Lane 2). Anti-Rabbit lgG (HRP), specific to the non-reduced form of lgG at 1/1500 was used as the secondary antibody.

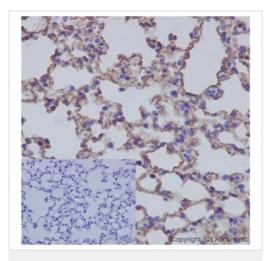
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab192869).



Immunocytochemistry/ Immunofluorescence - Anti-Caveolin-1 antibody [EPR15554] - BSA and Azide free (ab240332)

Immunocytochemistry/Immunofluorescence analysis of A-673 cells labelling Caveolin-1 with <u>ab192869</u> at a dilution of 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. <u>ab150077</u>, an Alexa Fluor[®] 488-conjugated goat antirabbit IgG (1/1000) was used as the secondary antibody. The cells were co-stained with <u>ab195889</u>, Alexa Fluor[®] 594-conjugated anti-Tubulin [DM1A] at a dilution of 1/200. Nuclei counterstained with DAPI (blue).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab192869).



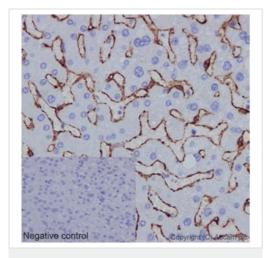
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Caveolin-1 antibody

[EPR15554] - BSA and Azide free (ab240332)

Immunohistochemical analysis of paraffin-embedded Mouse lung tissue labeling Caveolin-1 using <u>ab192869</u> at 1/4000 dilution (0.15 µg/ml). A prediluted HRP Polymer for Rabbit lgG was used as the secondary. Counterstain: Hematoxylin. Negative control also shown.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab192869</u>).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



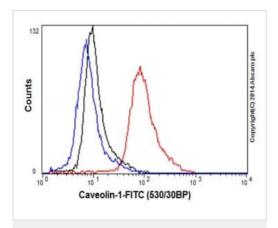
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Caveolin-1 antibody

[EPR15554] - BSA and Azide free (ab240332)

Immunohistochemical analysis of paraffin-embedded Human liver tissue labeling Caveolin-1 using <u>ab192869</u> at 1/4000 dilution (0.15 µg/ml). A prediluted HRP Polymer for Rabbit lgG was used as the secondary. Counterstain: Hematoxylin. Negative control also shown.

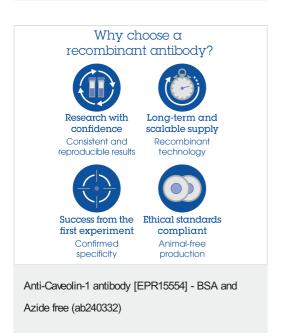
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab192869).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-Caveolin-1 antibody [EPR15554] - BSA and Azide free (ab240332) Intracellular Flow Cytometry analysis of NIH3T3 cells labeling Caveolin-1 using ab192869 at a 1/120 dilution (Red). A Goat anti rabbit lgG (FITC) at 1/150 dilution was used as secondary antibody. Cells were fixed with 2% paraformaldehyde. Cells without incubation with primary antibody and secondary antibody Blue. Rabbit monoclonal lgG was used as isotype control (Black).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab192869).



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