abcam

Product datasheet

Anti-Caveolin-1 antibody [E249] - Caveolae Marker ab32577





重组 RabMAb

★★★★★ 2 Abreviews 19 References 11 图像

概述

产品名称 Anti-Caveolin-1抗体[E249] - Caveolae Marker

描述 兔单克隆抗体[E249] to Caveolin-1 - Caveolae Marker

宿主 Rabbit

特异性 This antibody should recognize both alpha and beta form of Caveolin-1.

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

种属反应性 与反应: Mouse, Rat, Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: A431, A549 and HeLa cell lysates. ICC/IF: HeLa and Jurkat cells.

常规说明 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 monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

性能

形式 Liquid

存放说明 Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

存储溶液 pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

纯度 Protein A purified

克隆 单克隆 克隆编号 E249 同种型 lgG

The Abpromise guarantee

Abpromise™承诺保证使用ab32577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
IHC-P	****(1)	1/2000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. Use 0.01M Sodium Citrate Buffer, pH 6.0. For unpurified use at 1:250.
ICC/IF		1/50.
Flow Cyt		1/20.
WB	*** <u>*</u>	1/1000 - 1/10000. Predicted molecular weight: 20 kDa.

&M	A-A
	47IN

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-

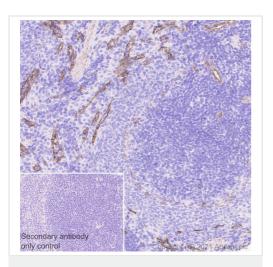
terminal amino acid is then N-acetylated.

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.

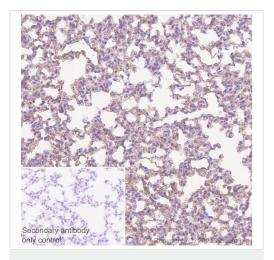
图片



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

[E249] - Caveolae Marker (ab32577)

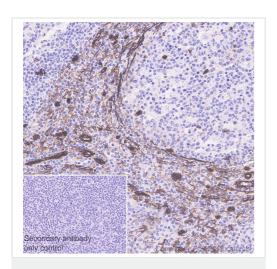
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat spleen tissue sections labelling Caveolin-1 with purified ab32577 at 1/2000 dilution (0.06 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



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

[E249] - Caveolae Marker (ab32577)

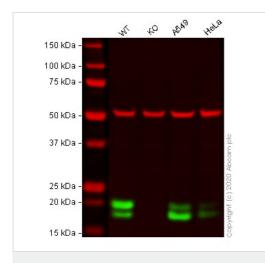
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse lung tissue sections labelling Caveolin-1 with purified ab32577 at 1/2000 dilution (0.06 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



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

[E249] - Caveolae Marker (ab32577)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue sections labelling Caveolin-1 with purified ab32577 at 1/2000 dilution (0.06 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Western blot - Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577)

All lanes : Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577) 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.

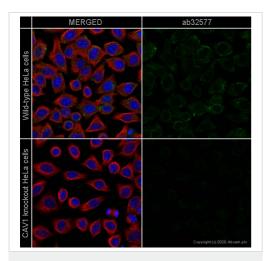
Performed under reducing conditions.

Predicted band size: 20 kDa Observed band size: 20 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab32577

observed at 20 kDa. Red - loading control, <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

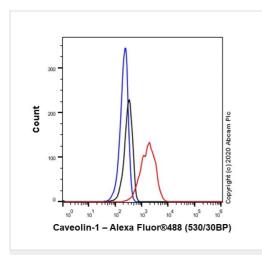
ab32577 was shown to react with Caveolin-1 in wild-type A431 cells in western blot. Loss of signal was observed when CAV1 knockout sample was used. A431 wild-type and CAV1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab32577 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.



Immunocytochemistry/ Immunofluorescence - Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577)

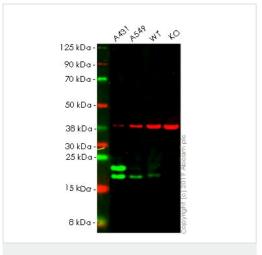
ab32577 staining Caveolin-1 in wild-type HeLa cells (top panel) and CAV1 knockout HeLa cells (ab255371) (bottom panel). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab32577 at 1/200 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).



Flow Cytometry - Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577)

Flow cytometric analysis of 4% Paraformaldehyde fixed 90% Methanol permeabilized A431 (Human epidermoid carcinoma epithelial cell) cells labelling Caveolin-1 with ab32577 at 1/20 dilution (10 µg/ml) compared with a Rabbit monoclonal lgG (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) at 1/2000 was used as the secondary antibody.



Western blot - Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577)

All lanes : Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577) at 1/1000 dilution

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

Lane 3: Wild-type HeLa cell lysate

Lane 4: CAV1 knockout HeLa cell lysate

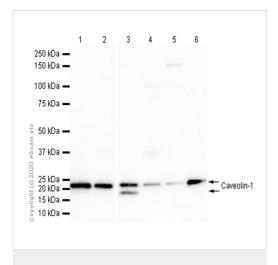
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 20 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab32577 observed at 20 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab32577 was shown to react with Caveolin-1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab255371 (knockout cell lysate ab263806) was used. Wild-type and Caveolin-1 knockout samples were subjected to SDS-PAGE. ab32577 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-Caveolin-1 antibody [E249] -Caveolae Marker (ab32577)

All lanes:

Lane 1: Mouse heart lysate

Lane 2: Mouse skeletal muscle lysate

Lane 3: Rat heart lysate

Lane 4: Rat skeletal muscle lysate

Lane 5 : C2C12 (Mouse myoblasts myoblast) whole cell lysate

Lane 6 : NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

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

Predicted band size: 20 kDa

Observed band size: 17, 23 kDa

The two isoforms of Caveolin-1 have been described in the literatures (PMID: 12816877, 11748292 and 14992406). We are unsure about the nature of the 250kDa bands.

All lanes: Anti-Caveolin-1 antibody [E249] - Caveolae Marker

(ab32577) at 1/1000 dilution (Purified)

Lane 1 : A431 (Human epidermoid carcinoma epithelial cell) whole cell lysate

Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 3: HaCaT (Human skin keratinocyte) whole cell lysate

Lysates/proteins at 20 µg per lane.

250 kDa — 150 kDa —

100 kDa —

Western blot - Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577)

Secondary

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

Predicted band size: 20 kDa

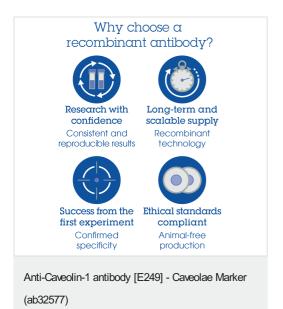
Observed band size: 17, 23 kDa

The two isoforms of Caveolin-1 have been described in the literatures (PMID: 12816877, 11748292 and 14992406). We are unsure about the nature of the 250kDa bands.

ab32577 MERGED

DAPI Secondary antibody only control

Immunocytochemistry/ Immunofluorescence - Anti-Caveolin-1 antibody [E249] - Caveolae Marker (ab32577) Immunocytochemistry analysis of Jurkat (Human lung carcinoma epithelial cell) cells labeling Caveolin-1 with purified ab32577 at 1/50 dilution (2.3 μg/mL). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) 1/200 (2.5 μg/mL). Goat anti rabbit lgG (Alexa Fluor[®] 488, ab150078) was used as the secondary antibody at 1/1000 (2 μg/mL) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



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