abcam

Product datasheet

Anti-M6PR (cation independent) antibody [EPR6599] ab124767





重组 RabMAb

★★★★★ 7 Abreviews 61 References 15 图像

概述

产品名称 Anti-M6PR (cation independent)抗体[EPR6599]

描述 兔单克隆抗体[EPR6599] to M6PR (cation independent)

宿主 Rabbit

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

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 Jurkat, 293T, C6, PC-12, NIH/3T3 and Caco-2 cell lysates; Human papillary carcinoma, thyroid

and tonsil tissue; Mouse heart, kidney, colon and spleen tissue; Rat colon tissue . ICC/IF: HAP1

WT and HAP1-IGF2R knockout cells

This product is a recombinant monoclonal antibody, which offers several advantages including: 常规说明

- 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**.

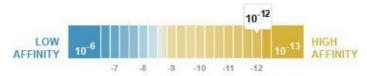
性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Stable for 12 months at -20°C.

 $K_D = 3.90 \times 10^{-12} M$ 解离常数(KD)



Learn more about K_D

存储溶液 pH: 7.20 Preservative: 0.01% Sodium azide

Constituents: 40% Glycerol, 0.05% BSA, 59% PBS

纯**度** Protein A purified

同种型 lgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab124767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
Flow Cyt (Intra)		1/100 - 1/500. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	**** <u>(4)</u>	1/50000 - 1/200000. Detects a band of approximately 300 kDa (predicted molecular weight: 274 kDa).
IHC-P		1/250 - 1/500. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. Heat up to 98°C, below boiling, and then let cool for 10-20 min. See IHC antigen retrieval protocols.
ICC/IF	★★★★★ (3)	Use a concentration of 1 µg/ml.

靶标

功能 Transport of phosphorylated lysosomal enzymes from the Golgi complex and the cell surface to

lysosomes. Lysosomal enzymes bearing phosphomannosyl residues bind specifically to

mannose-6-phosphate receptors in the Golgi apparatus and the resulting receptor-ligand complex is transported to an acidic prelyosomal compartment where the low pH mediates the dissociation of the complex. This receptor also binds IGF2. Acts as a positive regulator of T-cell coactivation,

by binding DPP4.

序列相似性 Belongs to the MRL1/IGF2R family.

Contains 1 fibronectin type-II domain.

结**构域** Contains 15 repeating units of approximately 147 AA harboring four disulfide bonds each. The

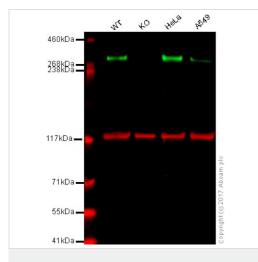
most highly conserved region within the repeat consists of a stretch of 13 AA that contains

cysteines at both ends.

细胞定位 Lysosome membrane. Colocalized with DPP4 in internalized cytoplasmic vesicles adjacent to the

cell surface.

图片



Western blot - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

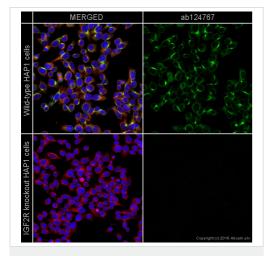
Lane 2: M6PR (cation independent) knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: A549 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab124767 observed at 274 kDa. Red - loading control, <u>ab18058</u>, observed at 130 kDa.

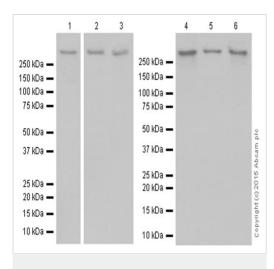
ab124767 was shown to specifically react with M6PR (cation independent) in wild-type HAP1 cells as signal was lost in M6PR (cation independent) knockout cells. Wild-type and M6PR (cation independent) knockout samples were subjected to SDS-PAGE. Ab124767 and ab18058 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/50000 dilution and 1/20000 dilution respectively. Blots were developed 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 1/10000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

ab124767 staining M6PR in wild-type HAP1 cells (top panel) and IGF2R knockout HAP1 cells (bottom panel). The cells were fixed with 100% MeOH for 5 min., 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 ab124767 at 1µg/ml and ab195889 (Mouse monoclonal [DM1A] to alpha Tubulin - Microtubule Marker (Alexa Fluor® 594)) at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor® 488) (ab150081) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

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



Western blot - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

All lanes : Anti-M6PR (cation independent) antibody [EPR6599] (ab124767) at 1/50000 dilution (purified)

Lane 1: C6 (rat glioma) whole cell lysate

Lane 2: PC-12 (rat adrenal gland pheochromocytoma) whole cell

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Lane 3: NIH/3T3 (mouse embryo) whole cell lysate

Lane 4 : Mouse heart tissue lysate
Lane 5 : Mouse kidney tissue lysate
Lane 6 : Mouse spleen tissue lysate

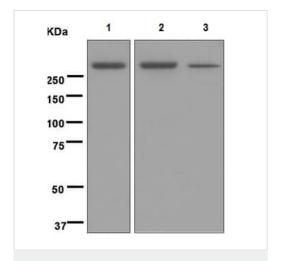
Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 274 kDa Observed band size: 300 kDa

Blocking and diluting buffer 5% NFDM/TBST



Western blot - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

All lanes : Anti-M6PR (cation independent) antibody [EPR6599] (ab124767) at 1/50000 dilution (unpurified)

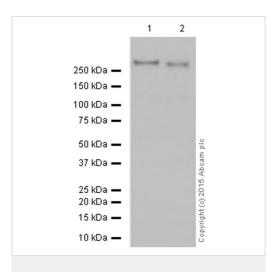
Lane 1 : Jurkat cell lysate
Lane 2 : 293T cell lysate
Lane 3 : Caco-2 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: Goat anti-Rabbit HRP at 1/2000 dilution

Predicted band size: 274 kDa



Western blot - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

All lanes : Anti-M6PR (cation independent) antibody [EPR6599] (ab124767) at 1/200000 dilution (purified)

Lane 1 : Jurkat (human acute T cell leukemia) whole cell lysate

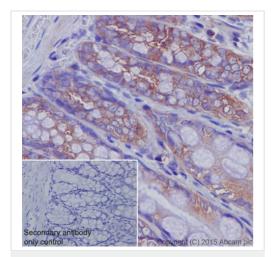
Lane 2 : HEK293 (human embryonic kidney) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

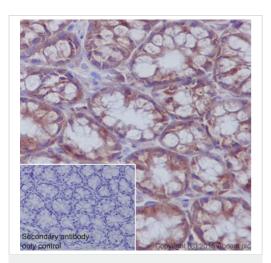
Predicted band size: 274 kDa Observed band size: 300 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

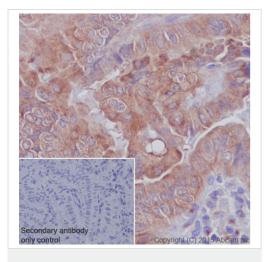
Blocking and diluting buffer 5% NFDM/TBST

Immunohistochemical staining of paraffin embedded rat colon tissue section labelling M6PR with purified ab124767 at dilution of 1/500. The secondary antibody used was ab97051 Goat Anti-Rabbit IgG H&L (HRP), at a dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.



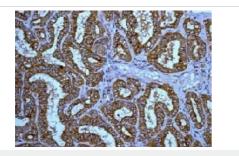
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

Immunohistochemical staining of paraffin embedded mouse colon tissue section labelling M6PR with purified ab124767 at dilution of 1/500. The secondary antibody used was ab97051 Goat Anti-Rabbit IgG H&L (HRP), at a dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

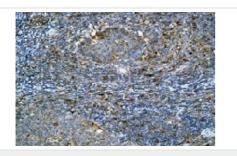
Immunohistochemical staining of paraffin embedded human thyroid carcinoma tissue section labelling M6PR with purified ab124767 at dilution of 1/500. The secondary antibody used was ab97051 Goat Anti-Rabbit IgG H&L (HRP), at a dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control and is shown in the inset.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

Immunohistochemical analysis of formalin fixed, paraffin embedded Human papillary carcinoma tissue section labelling Mannose 6 Phosphate Receptor (Cation independent) with unpurified ab124767 at dilution of 1/250.

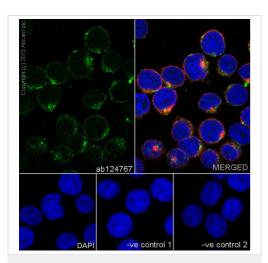
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

Immunohistochemical analysis of formalin fixed, paraffin embedded Human tonsil tissue section labelling Mannose 6 Phosphate Receptor (Cation independent) with unpurified ab124767 at dilution of 1/250.

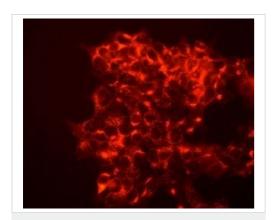
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

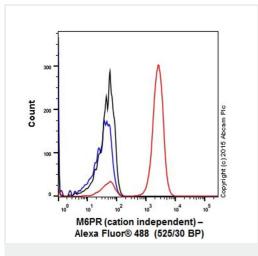
Immunocytochemistry/Immunofluorescence analysis of Jurkat (human acute T cell leukemia) cells labelling M6PR with purified ab124767 at 1/100. Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, Alexa Fluor[®] 488-conjugated goat anti-rabbit lgG (1/1000) was used as the secondary antibody. The cells were co-stained with ab7291, a mouse anti-tubulin antibody (1/1000) using ab150120, an Alexa Fluor[®] 594-conjugated goat anti-mouse lgG (1/1000) as the secondary. Nuclei couterstained with DAPI (blue).

For negative control 1, rabbit primary antibody was used, followed by anti-mouse secondary antibody (<u>ab150120</u>). For negative control 2, mouse primary antibody (<u>ab7291</u>) was used followed by anti-rabbit secondary antibody (<u>ab150077</u>).



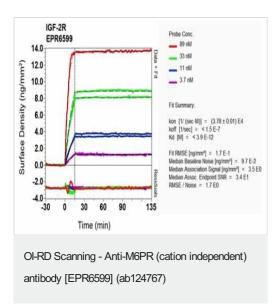
Immunocytochemistry/ Immunofluorescence - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

Immunocytochemistry/immunofluorescence analysis of 293T cells labelling Mannose 6 Phosphate Receptor (Cation independent) with unpurified ab124767 at dilution of 1/100.



Flow Cytometry (Intracellular) - Anti-M6PR (cation independent) antibody [EPR6599] (ab124767)

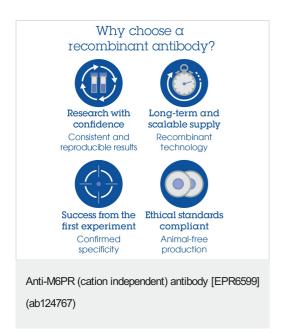
Intracellular Flow Cytometry analysis of Jurkat (human acute T cell leukemia) cells labelling M6PR with purified ab124767 at 1/150 (red). Cells were fixed with 4% paraformaldehyde. Alexa Fluorr® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



Equilibrium disassociation constant (K_D)

Click here to learn more about K_D

Learn more about KD



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