

Product datasheet

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

KO **VALIDATED** Recombinant RabMAb

★★★★★ 1 Abreviews 19 References 14 Images

Overview

Product name	Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker
Description	Rabbit monoclonal [EPR6599] to M6PR (cation independent) - Lysosome Membrane Marker
Host species	Rabbit
Tested applications	Suitable for: WB, IP, IHC-P, Flow Cyt, ICC/IF
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide within Human M6PR (cation independent) aa 2450 to the C-terminus (C terminal). The exact sequence is proprietary. Database link: P11717 (Peptide available as ab169957)
Positive control	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
General notes	

Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to [RabMab[®] patents](#).

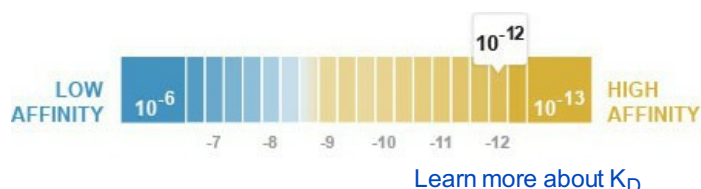
We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.

This product is a [recombinant rabbit monoclonal antibody](#).

Properties

Form	Liquid
Storage instructions	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.

Dissociation constant (K_D) K_D = 3.90 x 10⁻¹² M



Storage buffer pH: 7.20
Preservative: 0.01% Sodium azide
Constituents: 40% Glycerol, 0.05% BSA, 59% PBS

Purity Protein A purified

Clonality Monoclonal

Clone number EPR6599

Isotype IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab124767** 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	★★★★★	1/50000 - 1/200000. Detects a band of approximately 300 kDa (predicted molecular weight: 274 kDa). Can be blocked with M6PR (cation independent) peptide (ab169957) .
IP		1/10 - 1/100.
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 .
Flow Cyt		1/100 - 1/500. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		Use a concentration of 1 µg/ml.

Target

Function 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 prelysosomal 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.

Sequence similarities Belongs to the MRL1/IGF2R family.
Contains 1 fibronectin type-II domain.

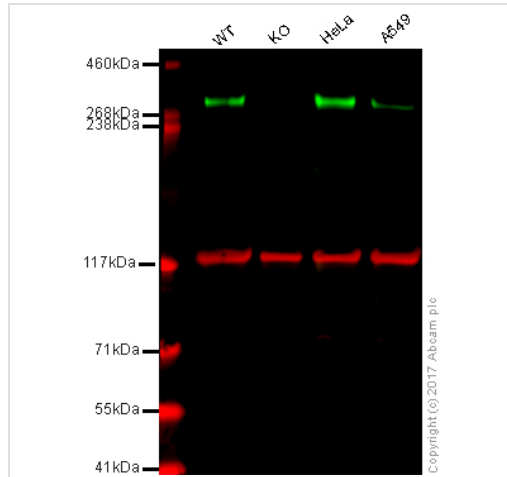
Domain Contains 15 repeating units of approximately 147 AA harboring four disulfide bonds each. The

most highly conserved region within the repeat consists of a stretch of 13 AA that contains cysteines at both ends.

Cellular localization

Lysosome membrane. Colocalized with DPP4 in internalized cytoplasmic vesicles adjacent to the cell surface.

Images



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

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

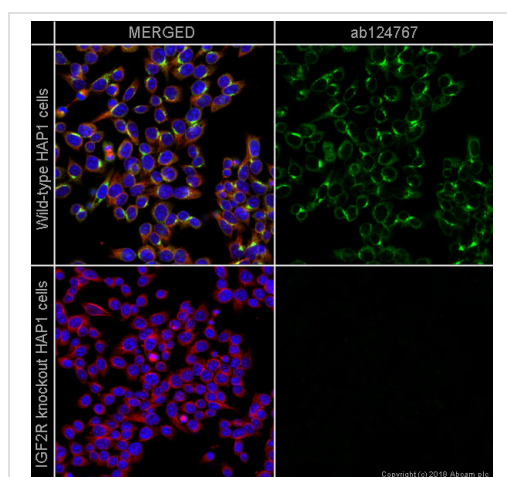
Lane 2: M6PR (cation independent) knockout HAP1 whole cell lysate (20 µg)

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, ab18058, observed at 130 kDa.

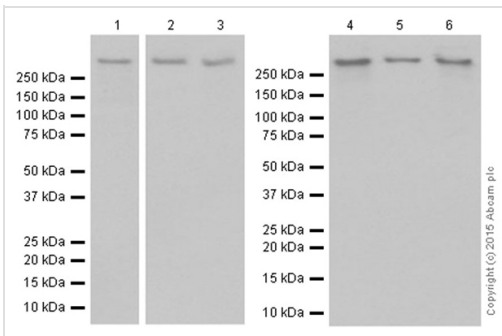
ab124767 was shown to specifically react with M6PR (cation independent) in wild-type HAP1 cells as signal was lost in M6PR (cation independent) knockout cells. Wild-type and M6PR (cation independent) knockout samples were subjected to SDS-PAGE. Ab124767 and ab18058 (Mouse anti-Vinculin loading control) 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] - Lysosome Membrane Marker (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 1 µg/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] - Lysosome Membrane Marker (ab124767)

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

Lane 1 : C6 (rat glioma) whole cell lysate

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

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

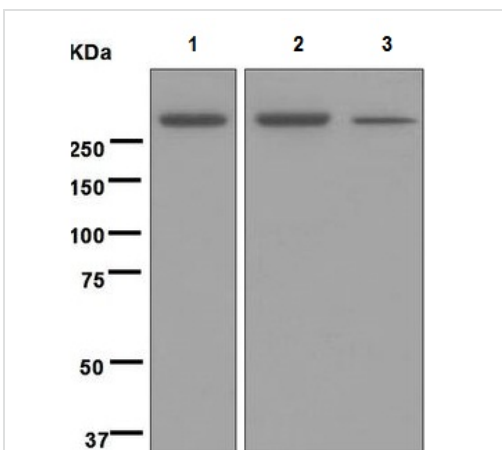
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 274 kDa

Observed band size: 300 kDa

[why is the actual band size different from the predicted?](#)

Blocking and diluting buffer 5% NFDm/TBST



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

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

Lane 1 : Jurkat cell lysate

Lane 2 : 293T cell lysate

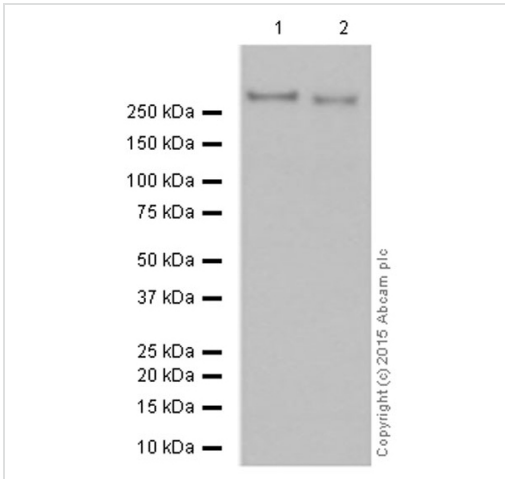
Lane 3 : Caco-2 cell lysate

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] - Lysosome Membrane Marker (ab124767)

All lanes : Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (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.

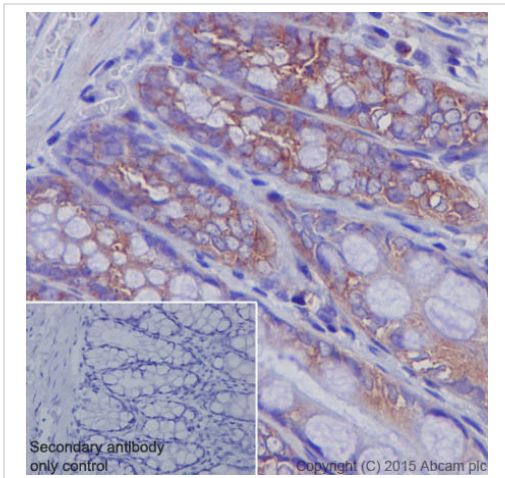
Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 274 kDa

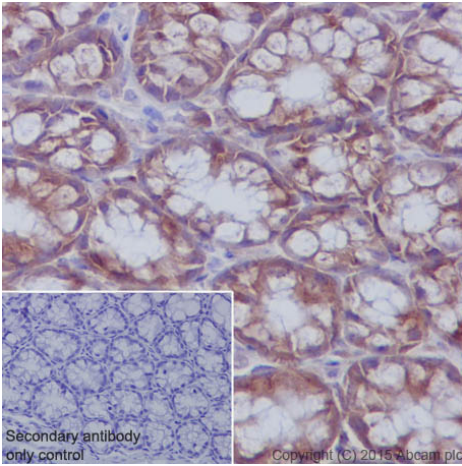
Observed band size: 300 kDa [why is the actual band size different from the predicted?](#)

Blocking and diluting buffer 5% NFDm/TBST



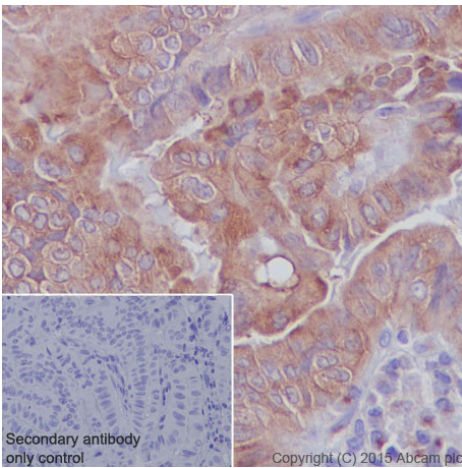
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)

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.



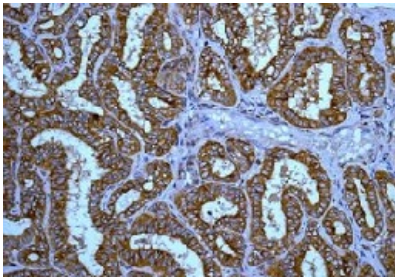
Immunohistochemical staining of paraffin embedded mouse colon tissue section labelling M6PR with purified ab124767 at dilution of 1/500. The secondary antibody used was [ab97051](#) Goat Anti-Rabbit IgG H&L (HRP), at a dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval 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 paraffin-embedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)



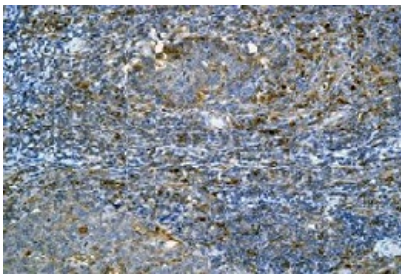
Immunohistochemical staining of paraffin embedded human thyroid carcinoma tissue section labelling M6PR with purified ab124767 at dilution of 1/500. The secondary antibody used was [ab97051](#) Goat Anti-Rabbit IgG H&L (HRP), at a dilution of 1/500. The sample was counter-stained with hematoxylin. Antigen retrieval was 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 paraffin-embedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)



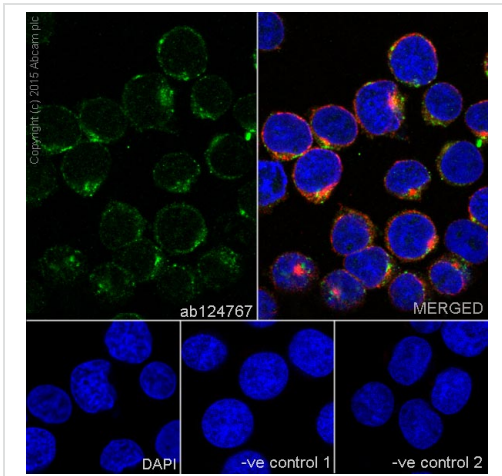
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)

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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)

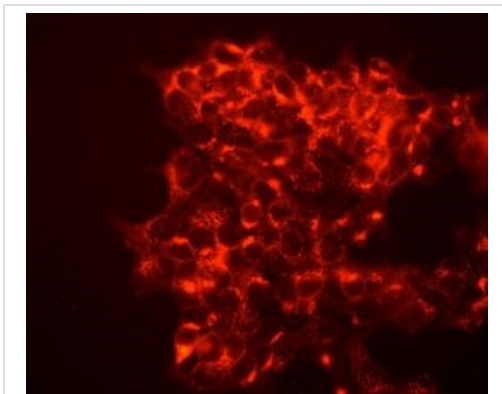
Immunohistochemical analysis of formalin fixed, paraffin embedded Human tonsil tissue section labelling Mannose 6 Phosphate Receptor (Cation independent) with unpurified ab124767 at dilution of 1/250.



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

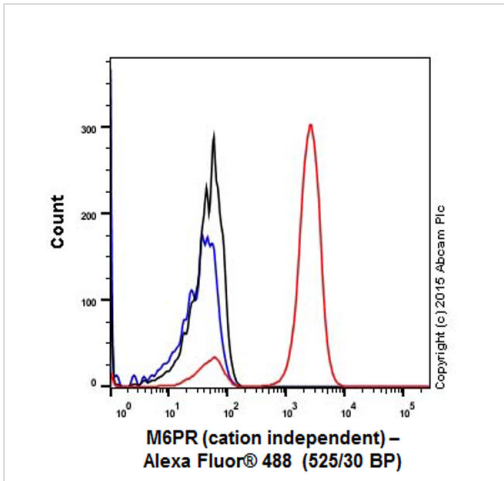
Immunocytochemistry/Immunofluorescence analysis of Jurkat (human acute T cell leukemia) cells labelling M6PR with purified ab124767 at 1/100. Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used 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 IgG (1/1000) as the secondary. Nuclei counterstained with DAPI (blue).

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



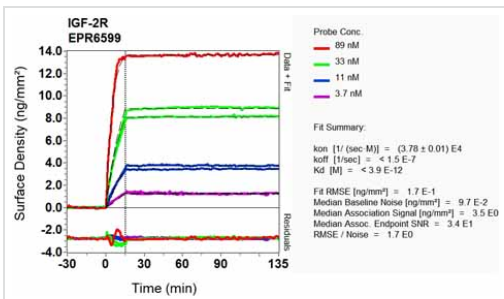
Immunocytochemistry/ Immunofluorescence - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)

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



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 Fluor® 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.

Flow Cytometry - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)



Equilibrium disassociation constant (K_D)

Learn more about K_D

[Click here to learn more about \$K_D\$](#)

Other - Anti-M6PR (cation independent) antibody [EPR6599] - Lysosome Membrane Marker (ab124767)

Please note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE"

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