

Anti-CD146 antibody [EPR3208] - BSA and Azide free ab210072

敲除验证
重组
RabMAb

[11 References](#)
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概述

产品名称	Anti-CD146抗体[EPR3208] - BSA and Azide free
描述	兔单克隆抗体[EPR3208] to CD146 - BSA and Azide free
宿主	Rabbit
经测试应用	适用于: Flow Cyt (Intra), WB, ICC/IF, IHC-P
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	ICC/IF: A375 cells. IHC-Fr: Mouse spleen tissue. IHC-P: Melanoma, breast carcinoma vessel, urinary bladder transitional carcinoma vessel, glioma vessel, normal tonsil and normal spleen tissue. WB: HeLa and HAP1 cell lysates. Flow Cyt (intra): A375 and HUVEC cells.
常规说明	<p>ab210072 is the carrier-free version of ab75769.</p> <p>Our carrier-free 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.</p> <p>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.</p> <p>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.</p> <p>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.</p> <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

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	EPR3208
同种型	IgG

应用

The Abpromise guarantee **Abpromise™**承诺保证使用ab210072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。

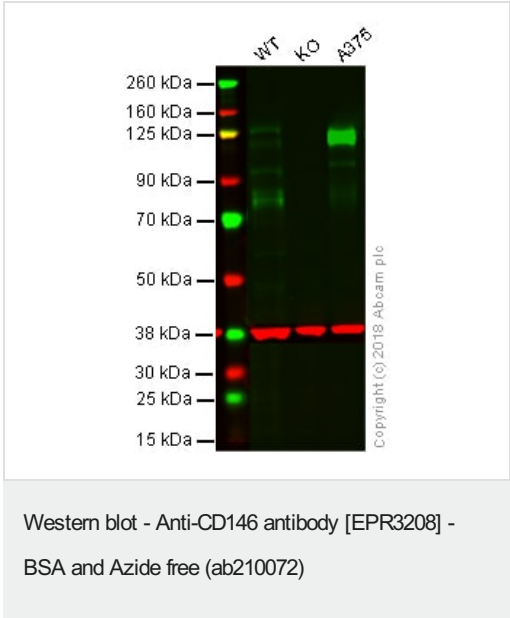
应用	Ab评论	说明
Flow Cyt (Intra)		Use at an assay dependent concentration. ab199376 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
WB		Use at an assay dependent concentration. Predicted molecular weight: 72 kDa.
ICC/IF		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol. See <u>IHC antigen retrieval protocols</u> .

靶标

功能	Plays a role in cell adhesion, and in cohesion of the endothelial monolayer at intercellular junctions in vascular tissue. Its expression may allow melanoma cells to interact with cellular elements of the vascular system, thereby enhancing hematogeneous tumor spread. Could be an adhesion molecule active in neural crest cells during embryonic development. Acts as surface receptor that triggers tyrosine phosphorylation of FYN and PTK2, and a transient increase in the intracellular calcium concentration.
组织特异性	Detected in endothelial cells in vascular tissue throughout the body. May appear at the surface of neural crest cells during their embryonic migration. Appears to be limited to vascular smooth muscle in normal adult tissues. Associated with tumor progression and the development of metastasis in human malignant melanoma. Expressed most strongly on metastatic lesions and advanced primary tumors and is only rarely detected in benign melanocytic nevi and thin primary melanomas with a low probability of metastasis.

序列相似性	Contains 3 Ig-like C2-type (immunoglobulin-like) domains. Contains 2 Ig-like V-type (immunoglobulin-like) domains.
细胞定位	Membrane.

图片



Lane 1: Wild-type HAP1 whole cell lysate (40 µg)

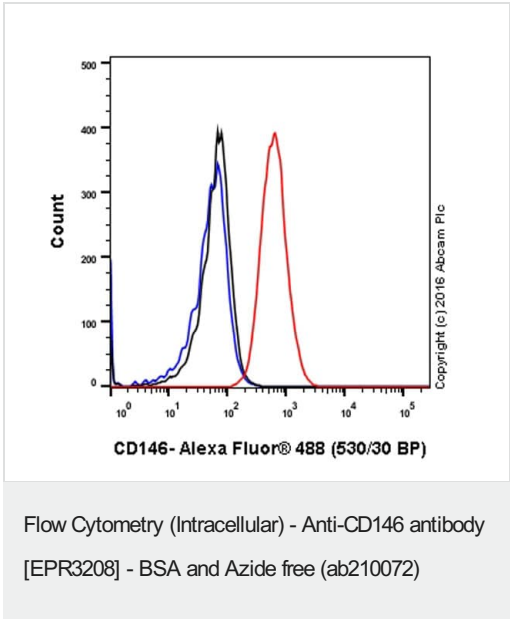
Lane 2: CD146 knockout HAP1 whole cell lysate (40 µg)

Lane 3: A375 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - [ab75769](#) observed at 120-72 kDa. Red - loading control, [ab9484](#), observed at 37 kDa.

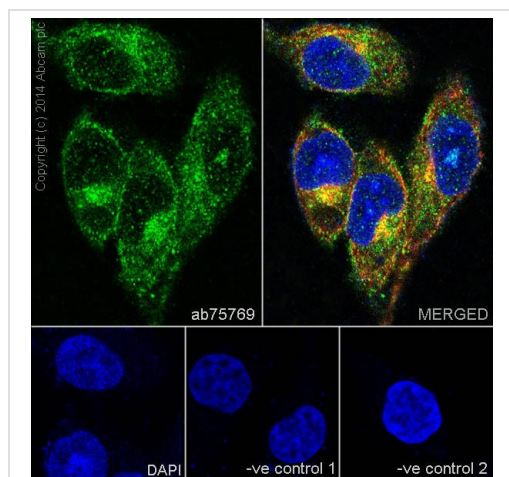
[ab75769](#) was shown to specifically react with CD146 in wild-type HAP1 cells as signal was lost in CD146 knockout cells. Wild-type and CD146 knockout samples were subjected to SDS-PAGE. [ab75769](#) and [ab9484](#) (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/10000 dilution 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab75769](#)).



Intracellular Flow Cytometry analysis of A375 (human malignant melanoma) cells labeling CD146 with unpurified [ab75769](#) at 1/20 dilution (10ug/mL) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor®488) at 1/2000 dilution was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) was used as the unlabeled control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab75769](#)).



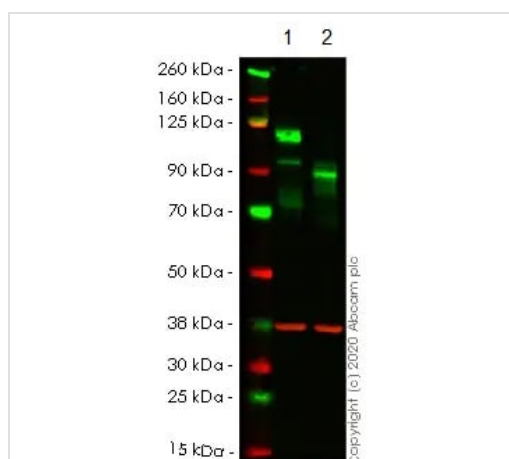
Immunocytochemistry/ Immunofluorescence - Anti-CD146 antibody [EPR3208] - BSA and Azide free (ab210072)

Immunocytochemistry/Immunofluorescence analysis of A375 (human malignant melanoma) cells labelling CD146 with purified **ab75769** at 1/250. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor[®] 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. **ab7291**, a mouse anti-tubulin (1/1000) and **ab150120**, an Alexa Fluor[®] 594-conjugated goat anti-mouse IgG (1/1000) were also used.

Control 1: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor[®] 594-conjugated goat anti-mouse IgG (1/500).

Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor[®] 488-conjugated goat anti-rabbit IgG (1/500).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75769**).



Western blot - Anti-CD146 antibody [EPR3208] - BSA and Azide free (ab210072)

All lanes : Anti-CD146 antibody [EPR3208] (**ab75769**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : MCAM CRISPR/Cas9 edited HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 72 kDa

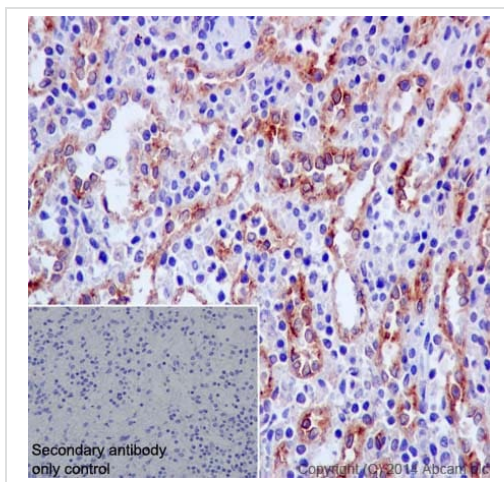
Observed band size: 120 kDa

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75769**).

Lanes 1-2: Merged signal (red and green). Green - **ab75769** observed at 120 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab75769 was shown to react with CD146 in wild-type HeLa cells in western blot. The band observed in CRISPR/Cas9 edited cell line

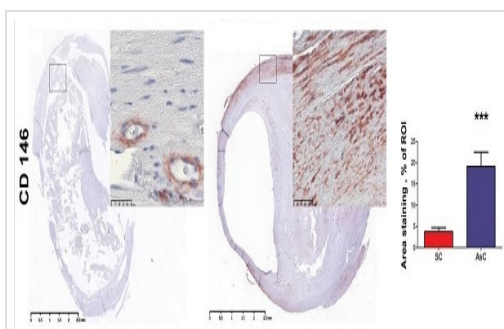
ab261790 (CRISPR/Cas9 edited cell lysate **ab256985**) lane below 120kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type HeLa and MCAM CRISPR/Cas9 edited HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab75769** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at a 1 in 1000 dilution and a 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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] - BSA and Azide free (**ab210072**)

This IHC data was generated using the same anti-CD146 antibody clone, EPR3208, in a different buffer formulation (cat# **ab75769**).

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human spleen tissue labelling CD146 with purified **ab75769** at 1/250. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a goat anti-rabbit IgG H&L (HRP) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] - BSA and Azide free (**ab210072**)

Image from Davaine J.M. et al. PLoS One. 2014 Sep 26;9(9):e107642. doi: 10.1371/journal.pone.0107642. eCollection 2014.

Immunohistochemistry experiments were used to compare symptomatic carotid plaques (SC) and asymptomatic carotid plaques (AsC)

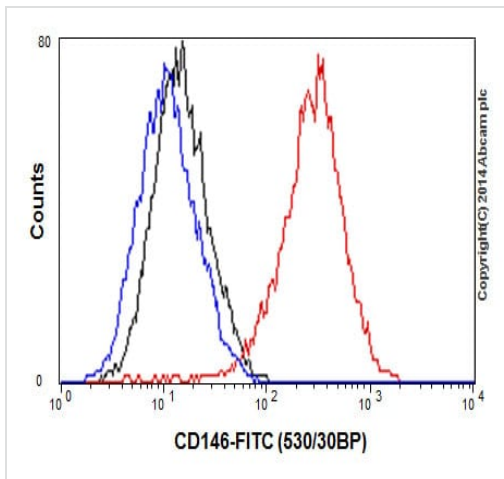
Asymptomatic lesions presented higher CD146⁺ pericyte infiltration, p

ab75769 used at 1/200 dilution.

(After Figure 2 of Davaine et al)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75769**).

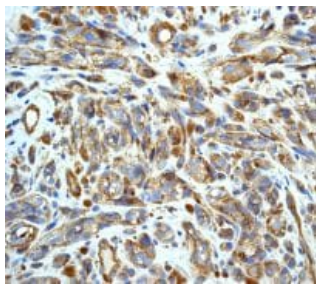
Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-CD146 antibody
[EPR3208] - BSA and Azide free (ab210072)

Intracellular Flow Cytometry analysis of HUVEC cells labelling CD146 with purified **ab75769** at 1/50 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75769**).

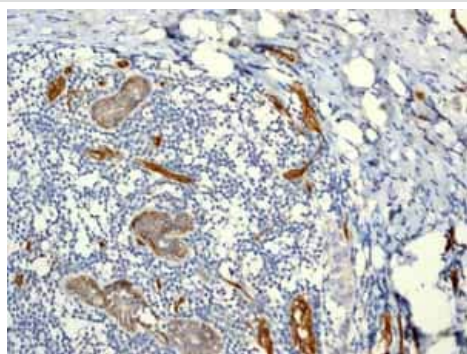


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody
[EPR3208] - BSA and Azide free (ab210072)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of melanoma tissue labelling CD146 with unpurified **ab75769** at 1/250. A HRP/AP polymerized secondary antibody was used.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75769**).

Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.

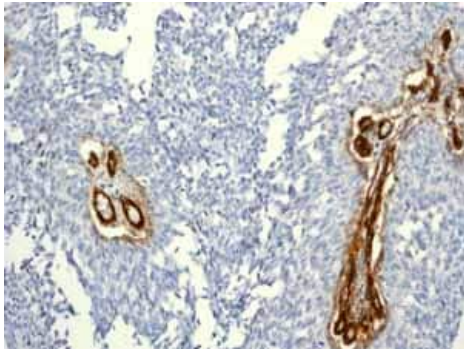


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody
[EPR3208] - BSA and Azide free (ab210072)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of breast carcinoma vessels tissue labelling CD146 with unpurified **ab75769**.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75769**).

Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.

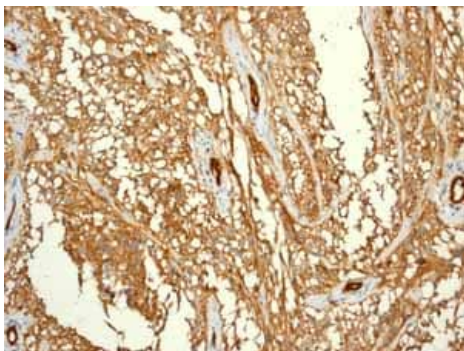


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] - BSA and Azide free (ab210072)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of urinary bladder transitional carcinoma vessels tissue labelling CD146 with unpurified [ab75769](#).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab75769](#)).

Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.

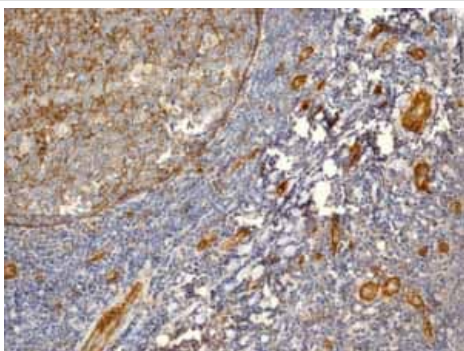


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] - BSA and Azide free (ab210072)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of glioma vessels tissue labelling CD146 with unpurified [ab75769](#).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab75769](#)).

Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.

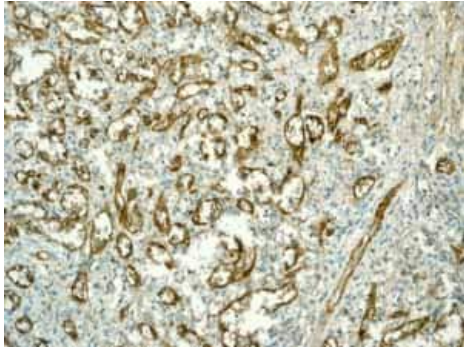


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] - BSA and Azide free (ab210072)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of normal tonsil tissue labelling CD146 with unpurified [ab75769](#).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab75769](#)).

Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.



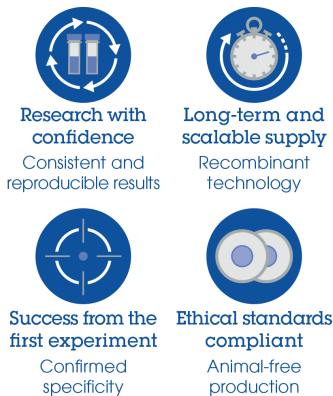
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] - BSA and Azide free (ab210072)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of normal spleen tissue labelling CD146 with unpurified **ab75769**.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab75769**).

Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.

Why choose a recombinant antibody?



Anti-CD146 antibody [EPR3208] - BSA and Azide free (ab210072)

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