abcam

ab75769

Anti-CD146 antibody [EPR3208]

Anti-CD146 antibody [EPR3208] (ab75769) is a rabbit monoclonal antibody detecting CD146 in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF**. Suitable for **Human, Mouse, Rat**.

- KO validated for confirmed specificity
- Biophysical QC for unrivalled batch-batch consistency
- Over 140 publications
- Trusted since 2009

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR3208
Purification technique	Affinity purification Protein A
Concentration	0.112 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/250 - 1/500
Notes	Perform heat-mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Mouse
Dilution info	1/1000
Notes	**For unpurified use at 1/10000 - 1/50000.**

Species	Rat
Dilution info	1/1000
Notes	**For unpurified use at 1/10000 - 1/50000.**

Species	Human
Dilution info	1/1000
Notes	**For unpurified use at 1/10000 - 1/50000.**

ICC/IF

Tested

Species	Human
Dilution info	1/100 - 1/250
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/20 - 1/80
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

IHC-Fr

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

Target data

See full target information MCAM

Function	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 a surface receptor that triggers tyrosine phosphorylation of FYN and PTK2/FAK1, and a transient increase in the intracellular calcium concentration.

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Anti-CD146 antibody [EPR3208] (ab75769) was developed by Abcam using patented rabbit monoclonal antibody technology and is validated for use in Flow Cyt (Intra), ICC/IF, IHC-P, WB in human, mouse, rat samples.

Anti-CD146 antibody [EPR3208] (ab75769) has been cited over 149 times in peer reviewed journals and is trusted by the scientific community.

Abcam's high quality manufacturing and validation processes ensure Anti-CD146 antibody [EPR3208] (ab75769) has high sensitivity and specificity alongside high lot-to-lot consistency and reproducibility.

The specificity of Anti-CD146 antibody [EPR3208] (ab75769) has been confirmed by testing in knockout samples.

Anti-CD146 antibody [EPR3208] (ab75769) has 40 independent reviews from customers.

Anti-CD146 antibody [EPR3208] (ab75769) specifically detects CD146 (UniProt ID: P43121; Molecular weight: 69kDa) and is sold in 100 µL and 1 mL selling sizes.

Conjugation-ready, carrier free format available for antibody clone EPR3208 - ab210072.

Antibody clone EPR3208 is also available pre-conjugated to a variety of labels for your convenience - Alexa Fluor® 488, PE, PerCP, Biotin, APC, HRP, Alexa Fluor® 647, Alexa Fluor® 555 (ab196448, ab209298, ab216361, ab217072, ab303023, ab303024, ab305213, ab307390).

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	CD146 also known as MCAM or MUC18 is a cell adhesion molecule that belongs to the immunoglobulin superfamily. With a mass of approximately 113 kDa CD146 is expressed mainly on the surface of endothelial cells but it can also be found on smooth muscle cells some T-cells and in certain cancerous tissues. This protein plays a role in cell-cell adhesion contributing to the integrity and signaling functions within tissue microenvironments.
Biological function summary	CD146 influences the migration and organization of cells within the vascular system. It serves as a component of cell adhesion complexes facilitating interactions between endothelial cells and other cell types. Additionally CD146 contributes to angiogenesis the process by which new blood vessels form from existing vasculature which is critical during tissue development and repair. This function makes it an important element in maintaining normal physiological processes.
Pathways	CD146 participates in the Wnt/β-catenin and MAPK signaling pathways which are vital in cell proliferation and differentiation. Through the Wnt/β-catenin pathway CD146 interacts with other proteins like Frizzled receptors to regulate gene transcription. In the MAPK pathway it is linked with signaling cascades that involve proteins such as ERK leading to cellular responses necessary for growth and response to external stimuli.
Associated diseases and disorders	CD146 has been implicated in melanoma progression and cardiovascular diseases. Its overexpression in melanoma associates it with increased tumor growth and metastasis often involving interactions with other adhesion molecules like integrins. In cardiovascular diseases CD146 relates to atherosclerosis as its expression on endothelial cells can promote inflammatory responses. Understanding these connections provides insights into potential therapeutic targets for treating these conditions.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

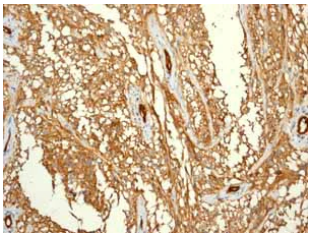
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

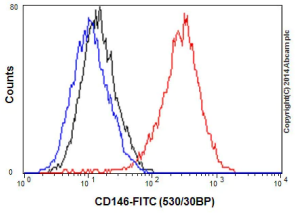
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19 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

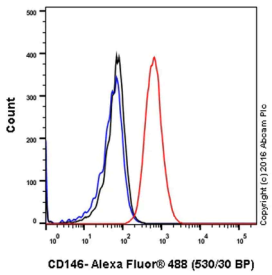
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of glioma vessels tissue labelling CD146 with unpurified ab75769.
Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Flow Cytometry (Intracellular) staining using rabbit Anti-CD146 antibody

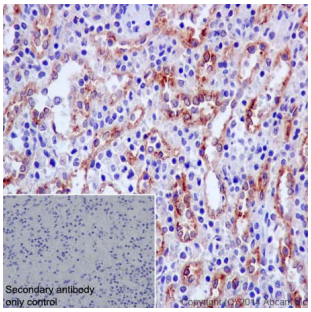
Intracellular Flow Cytometry analysis ofHUVEC cells labelling CD146 with purified ab75769 at 1/50 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control,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



Flow Cytometry (Intracellular) - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Flow Cytometry (Intracellular) staining using rabbit Anti-CD146 antibody

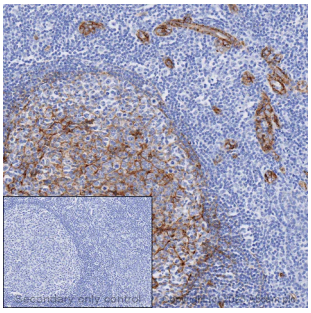
Intracellular Flow Cytometry analysis of A375 (human malignant melanoma) cells labeling CD146 with unpurified ab75769 at 1/20 dilution (10ug/mL) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluorr®488) at 1/2000 dilution was used 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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD146 antibody

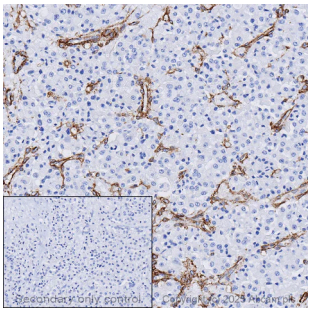
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human spleen tissue labelling CD146 with purified ab75769 at 1/250. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. ab97051, a goat anti-rabbit IgG H&L (HRP) was used as the secondary antibody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



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human tonsil using rabbit Anti-CD146 antibody

Immunohistochemical analysis of formalin fixed paraffin embedded human tonsil labelling CD146 with ab75769 at a concentration of 0.1µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with a OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was performed with DISCOVERY cell conditioning solution (CC1) 100°C, pH8.5 for 32mins. ab75769 Anti-CD146 antibody [EPR3208] was incubated for 16mins at 37°C. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.
Customers are encouraged to optimise antigen retrieval conditions, antibody concentration, incubation times and temperature for best results in their own IHC assay workflow (automated and manual).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human liver hepatocellular carcinoma using rabbit Anti-CD146 antibody

Immunohistochemical analysis of formalin fixed paraffin embedded human liver hepatocellular carcinoma labelling CD146 with ab75769 at a concentration of 0.1µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with a OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was performed with DISCOVERY cell

conditioning solution (CC1) 100°C, pH8.5 for 32mins. ab75769 Anti-CD146 antibody [EPR3208] was incubated for 16mins at 37°C. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.
Customers are encouraged to optimise antigen retrieval conditions, antibody concentration, incubation times and temperature for best results in their own IHC assay workflow (automated and manual).

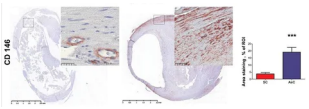


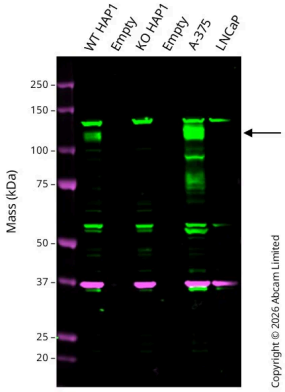
Image from Davaine J.M. et al. PLoS One. 2014 Sep 26;9(9):e107642. doi: 10.1371/journal.pone.0107642. eCollection 2014.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

Immunohistochemistry experiments were used to compare symptomatic carotid plaques (SC) and asymptomatic carotid plaques (AsC)
Asymptomatic lesions presented higher CD146^{+} pericyte infiltration, p<0.001. Representative images are on the left with corresponding quantification on the right.

ab75769 used at 1/200 dilution.

(After Figure 2 of Davaine et al)



Western blot - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Western blot staining using rabbit Anti-CD146 antibody

Western blot: Anti-CD146 antibody [EPR3208] ab75769 staining at 1/1000 dilution, shown in green; Mouse anti GAPDH ab8245 loading control staining at 1/20000 dilution, shown in magenta. Samples were not boiled before loading onto the gel. A band was observed at 120 kDa in Wild-type HAP1 Not Boiled cell line with no signal observed at this size in MCAM knockout HAP1 Not Boiled cell line. To generate this image, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 5% Milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit 800CW and Goat anti-Mouse 680RD at 1/20,000 dilution.
All lanes:
Western blot - Anti-CD146 antibody [EPR3208] (ab75769) at 1/1000 dilution

Lane 1:
Wild-type HAP1 Not Boiled cell lysates at 40 µg

Lanes 2 and 4:
Empty

Lane 3:
MCAM knockout HAP1 Not Boiled cell lysates at 40 µg

Lane 5:
A-375 Not Boiled cell lysates at 20 µg

Lane 6:
LNCaP Not Boiled cell lysates at 20 µg

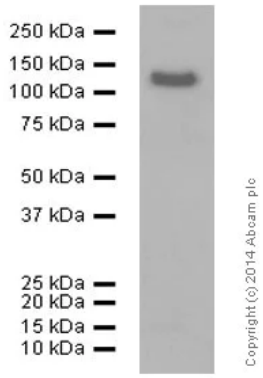
Secondary

All lanes:
Goat anti-Rabbit 800CW & Goat anti-Mouse 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 72 kDa

Observed band size: 120 kDa



Western blot - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Western blot staining of HUVEC cell lysate using rabbit Anti-CD146 antibody

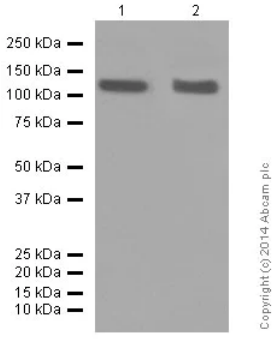
Blocking buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM /TBST.
All lanes:
Western blot - Anti-CD146 antibody [EPR3208] (ab75769) at 1/10000 dilution

All lanes:
HUVEC cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/1000 dilution

Predicted band size: 72 kDa



Western blot - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Western blot staining using rabbit Anti-CD146 antibody

Blocking buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM /TBST.
All lanes:
Western blot - Anti-CD146 antibody [EPR3208] (ab75769) at 1/10000 dilution

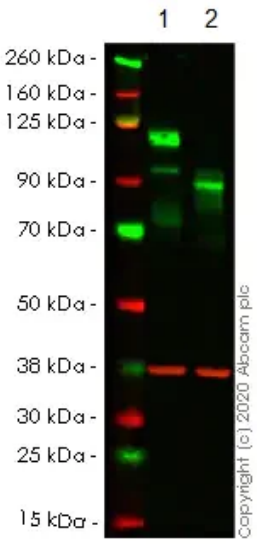
Lane 1:
A375 cell lysate at 20 µg

Lane 2:
Human fetal artery lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/1000 dilution

Predicted band size: 72 kDa



Western blot - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Western blot staining using rabbit Anti-CD146 antibody

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.

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

Lane 1:
Wild-type HeLa cell lysate at 20 µg

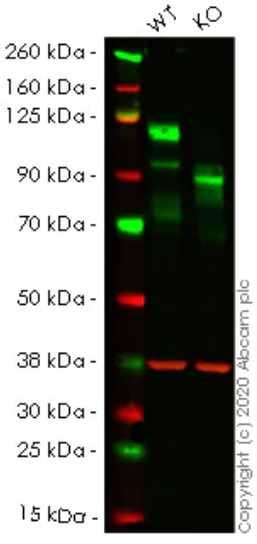
Lane 2:
MCAM CRISPR/Cas9 edited HeLa cell lysate at 20 µg

Lane 2:
Western blot - Human MCAM (CD146) knockout HeLa cell line (ab261790)

Performed under reducing conditions.

Predicted band size: 72 kDa

Observed band size: 120 kDa



Western blot - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Western blot staining using rabbit Anti-CD146 antibody

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 knockout cell line ab261790 (knockout cell lysate ab256985) lane below 120kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type HeLa and MCAM knockout 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.

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

Lane 1:
Wild-type HeLa cell lysate at 20 µg

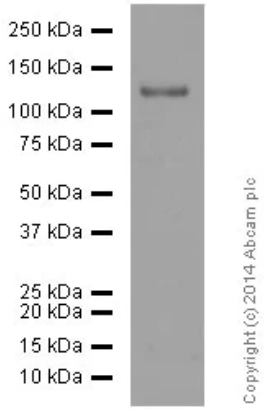
Lane 2:

MCAM knockout HeLa cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 72 kDa

Observed band size: 120 kDa



Western blot - Anti-CD146 antibody [EPR3208] (ab75769)

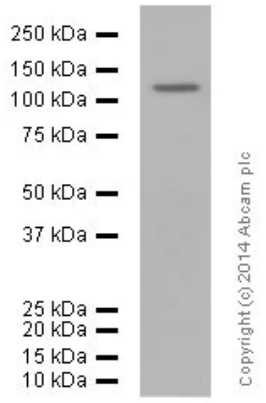
Blocking buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM /TBST.
All lanes:
Western blot - Anti-CD146 antibody [EPR3208] (ab75769) at 1/10000 dilution

All lanes:
Rat placenta lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/1000 dilution

Predicted band size: 72 kDa



Western blot - Anti-CD146 antibody [EPR3208] (ab75769)

CD146 Western blot staining of B16-F0 cell lysate using rabbit Anti-CD146 antibody

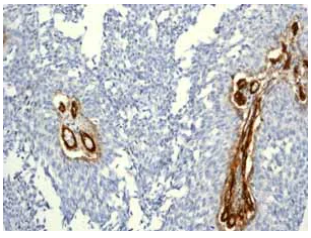
Blocking buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM /TBST.
All lanes:
Western blot - Anti-CD146 antibody [EPR3208] (ab75769) at 1/10000 dilution

All lanes:
B16-F0 cell lysate at 20 µg/mL

Secondary

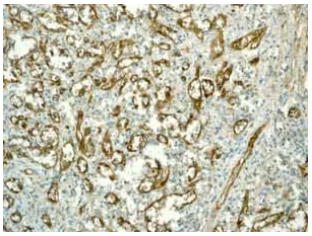
All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/1000 dilution

Predicted band size: 72 kDa



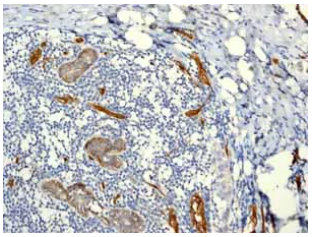
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of urinary bladder transitional carcinoma vessels tissue labelling CD146 with unpurified ab75769.
Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.



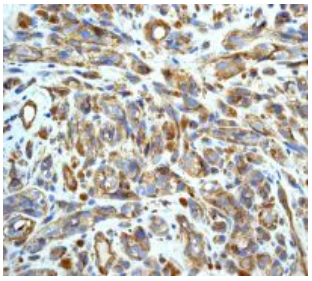
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of normal spleen tissue labelling CD146 with unpurified ab75769.
Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.



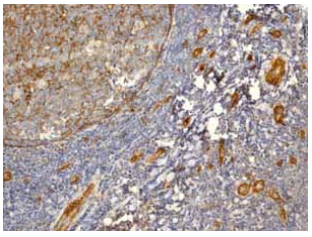
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD146 antibody [EPR3208] (ab75769)

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

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.
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] (ab75769)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of normal tonsil tissue labelling CD146 with unpurified ab75769.
Perform heat mediated antigen retrieval via the pressure cooker method before commencing with IHC staining protocol.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.