# abcam

#### Product datasheet

## Anti-CD147 antibody [EPR4053] ab108308





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#### Overview

**Product name** Anti-CD147 antibody [EPR4053]

**Description** Rabbit monoclonal [EPR4053] to CD147

**Host species** Rabbit

**Tested applications** Suitable for: WB, IHC-P, Flow Cyt, ICC/IF

Species reactivity Reacts with: Human

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: A549, Raji, HEK-293T, Jurkat, HuT-78, A431, U-87 MG and HeLa cell lysates; IHC-P:

Human colon cancer tissue; ICC/IF: HeLa cells; Flow Cyt: HeLa cells. WB: HeLa and U-87 MG

whole cell lysate.

**General notes** 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**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

## **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 long

term. Avoid freeze / thaw cycle.

pH: 7.20 Storage buffer

Preservative: 0.01% Sodium azide

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

Purity Protein A purified

ClonalityMonoclonalClone numberEPR4053IsotypeIgG

## **Applications**

The Abpromise guarantee

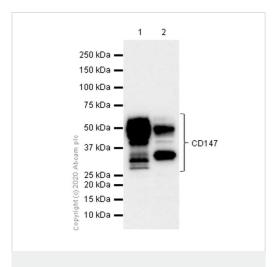
Our <u>Abpromise guarantee</u> covers the use of ab108308 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/1000 - 1/10000. Predicted molecular weight: 42 kDa.
IHC-P	<b>★★★☆☆ (1)</b>	1/3000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.  For unpurified use at 1/250 - 1/500.
Flow Cyt		1/30.
ICC/IF		1/50. For unpurified use at 1/500.

Target		
Function	Plays pivotal roles in spermatogenesis, embryo implantation, neural network formation and tumor progression. Stimulates adjacent fibroblasts to produce matrix metalloproteinases (MMPS). May target monocarboxylate transporters SLC16A1, SLC16A3 and SLC16A8 to plasma membranes of retinal pigment epithelium and neural retina. Seems to be a receptor for oligomannosidic glycans. In vitro, promotes outgrowth of astrocytic processes.	
Tissue specificity	Present only in vascular endothelium in non-neoplastic regions of the brain, whereas it is present in tumor cells but not in proliferating blood vessels in malignant gliomas.	
Sequence similarities	Contains 1 lg-like C2-type (immunoglobulin-like) domain. Contains 1 lg-like V-type (immunoglobulin-like) domain.	
Post-translational modifications	N-glycosylated.	
Cellular localization	Cell membrane. Melanosome. Colocalizes with SLC16A1 and SLC16A8 (By similarity). Identified by mass spectrometry in melanosome fractions from stage I to stage IV.	

## **Images**



Western blot - Anti-CD147 antibody [EPR4053] (ab108308)

**All lanes :** Anti-CD147 antibody [EPR4053] (ab108308) at 1/1000 dilution (Purified)

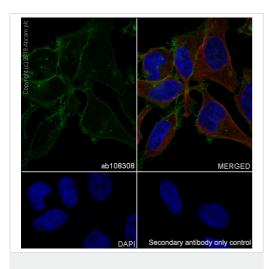
**Lane 1 :** HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

**Lane 2**: U-87 MG (Human glioblastoma-astrocytoma epithelial cell) whole cell lysate

### **Secondary**

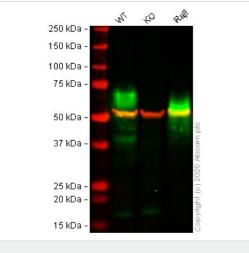
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 42 kDa



Immunocytochemistry/ Immunofluorescence - Anti-CD147 antibody [EPR4053] (ab108308)

Immunocytochemistry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling CD147 with purified ab108308 at 1:50 dilution (6.3 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 ,ab150077) 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.



Western blot - Anti-CD147 antibody [EPR4053] (ab108308)

**All lanes :** Anti-CD147 antibody [EPR4053] (ab108308) at 1/1000 dilution

Lane 1: Wild-type A549 cell lysate

Lane 2: BSG knockout A549 cell lysate

Lane 3: Raji cell lysate

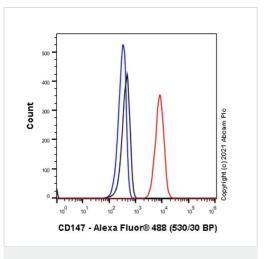
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 42 kDa **Observed band size:** 42-70 kDa

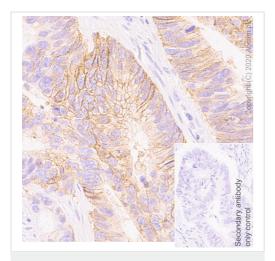
**Lanes 1 - 3:** Merged signal (red and green). Green - ab108308 observed at 42-70 kDa. Red - loading control <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

ab108308 was shown to react with CD147 in wild-type A549 cells in western blot with loss of signal observed in BSG knockout cell line <a href="mailto:ab273748">ab273748</a> (knockout cell lysate <a href="mailto:ab275500">ab275500</a>). Wild-type and BSG knockout A549 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with ab108308 and <a href="mailto:ab7291">ab7291</a> (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 lgG H&L (IRDye® 800CW) preabsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Flow Cytometry - Anti-CD147 antibody [EPR4053] (ab108308)

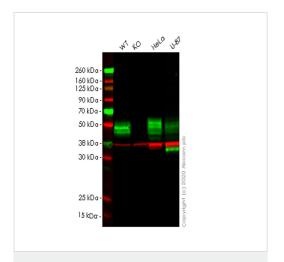
Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling CD147 with purified ab108308 at 1/30 dilution (10 µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, <u>ab150077</u>) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD147 antibody
[EPR4053] (ab108308)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon cancer tissue sections labeling CD147 with purified ab108308 at 1/3000 dilution (0.1 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 (<u>ab209101</u>) 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-CD147 antibody [EPR4053] (ab108308)

**All lanes :** Anti-CD147 antibody [EPR4053] (ab108308) at 1/1000 dilution

Lane 1: Wild-type HEK293T cell lysate

Lane 2: BSG knockout HEK293T cell lysate

Lane 3 : HeLa cell lysate

Lane 4 : U-87 MG cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

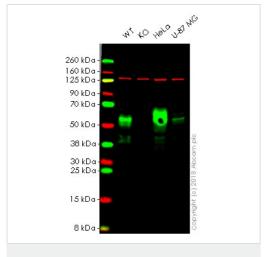
**All lanes :** Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Performed under reducing conditions.

**Predicted band size:** 42 kDa **Observed band size:** 50 kDa

**Lanes 1-4:** Merged signal (red and green). Green - ab108308 observed at 50 kDa. Red - loading control **ab8245** observed at 36 kDa.

ab108308 Anti-CD147 antibody [EPR4053] was shown to specifically react with CD147 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line <a href="mailto:ab266331">ab266331</a> (knockout cell lysate <a href="mailto:ab256853">ab256853</a>) was used. Wild-type and CD147 knockout samples were subjected to SDS-PAGE. ab108308 and Anti-GAPDH antibody [6C5] - Loading Control (<a href="mailto:ab8245">ab8245</a>) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-CD147 antibody [EPR4053] (ab108308)

**All lanes :** Anti-CD147 antibody [EPR4053] (ab108308) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: BSG (Basigin) knockout HAP1 whole cell lysate

Lane 3: HeLa whole cell lysate

Lane 4: U-87 MG whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 42 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab108308 observed at 42 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab108308 was shown to specifically react with Basigin in wild-type HAP1 cells as signal was lost in BSG (Basigin) knockout cells.

Wild-type and BSG (Basigin) knockout samples were subjected to SDS-PAGE. Ab108308 and <u>ab18058</u> (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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