# abcam

### Product datasheet

## Anti-CD147 antibody [EPR4053] - BSA and Azide free ab247626



Recombinant

RabMAb

### 7 Images

#### Overview

**Product name** Anti-CD147 antibody [EPR4053] - BSA and Azide free

**Description** Rabbit monoclonal [EPR4053] to CD147 - BSA and Azide free

**Host species** Rabbit

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

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. ICC/IF: HeLa

cells. IHC-P: Human colon cancer tissue Flow Cyt: HeLa cells. WB: HeLa and U-87 MG whole cell

lysate.

General notes ab247626 is the carrier-free version of ab108308.

> 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.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

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.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.

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

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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. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

**Purity** Protein A purified

Clonality Monoclonal
Clone number EPR4053

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab247626 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
ICC/IF		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.  See IHC antigen retrieval protocols.
WB		Use at an assay dependent concentration. Predicted molecular weight: 42 kDa.
Flow Cyt		Use at an assay dependent concentration.

#### **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.

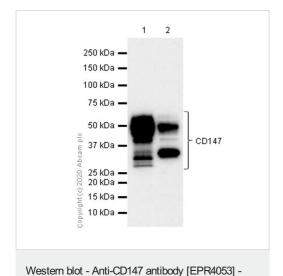
Post-translational modifications

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**



BSA and Azide free (ab247626)

**All lanes :** Anti-CD147 antibody [EPR4053] (<u>ab108308</u>) 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

dilution

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

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

Predicted band size: 42 kDa

250 kDa 150 kDa 150 kDa 75 kDa 37 kDa 25 kDa 20 kDa 15 kDa -

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

Western blot - Anti-CD147 antibody [EPR4053] -BSA and Azide free (ab247626)

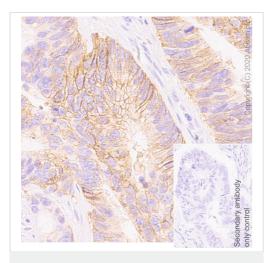
This data was developed using the same antibody clone in a different buffer formulation (ab108308).

Lanes 1 - 3: Merged signal (red and green). Green - <u>ab108308</u> 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 ab273748 (knockout cell lysate ab275500). 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 ab7291 (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 IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



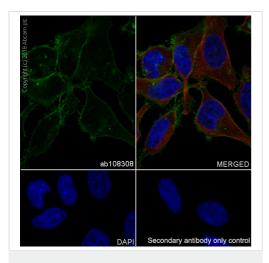
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD147 antibody

[EPR4053] - BSA and Azide free (ab247626)

This data was developed using <u>ab108308</u>, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon cancer tissue sections labeling CD147 with purified <u>ab108308</u> 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.

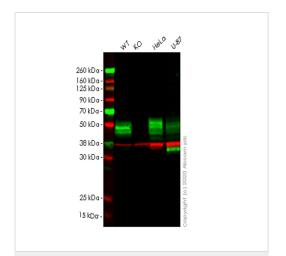
The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Immunocytochemistry/ Immunofluorescence - Anti-CD147 antibody [EPR4053] - BSA and Azide free (ab247626)

This data was developed using **ab108308**, the same antibody clone in a different buffer formulation.

Immunocytochemistry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling CD147 with purified  $\underline{ab108308}$  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  $^{\mbox{\scriptsize (B)}}$  594) 1/200 (2.5 µg/mL). Goat anti rabbit lgG (Alexa Fluor  $^{\mbox{\scriptsize (B)}}$  488,  $\underline{ab150077}$ ) 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] - BSA and Azide free (ab247626)

**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 (<u>ab216773</u>) 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 - <u>ab108308</u> observed at 50 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

<u>ab108308</u> 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 <u>ab266331</u> (knockout

cell lysate <u>ab256853</u>) was used. Wild-type and CD147 knockout samples were subjected to SDS-PAGE. <u>ab108308</u> and Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) 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 (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

CD147 - Alexa Fluor® 488 (530/30 BP)

Flow Cytometry - Anti-CD147 antibody [EPR4053] - BSA and Azide free (ab247626)

clone in a different buffer formulation.

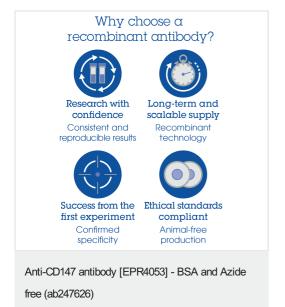
Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling CD147 with purified <u>ab108308</u> at 1/30 dilution (10 µg/mL) (Red). Cells were fixed with 4%

Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor® 488, <u>ab150077</u>) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG

(Black). Unlabelled control - Cell without incubation with primary

antibody and secondary antibody (Blue).

This data was developed using ab108308, the same antibody



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