abcam

ab666

# Anti-CD147 antibody [MEM-M6/1]

KO Validated

## Key facts

| | |
|---|---|
| **Isotype** | IgG1 |
| **Host species** | Mouse |
| **Storage buffer** | pH: 7.4 Preservative: 0.097% Sodium azide Constituents: PBS |
| **Form** | Liquid |
| **Clonality** | Monoclonal |
| **Immunogen** | Recombinant Fragment Protein within Human BSG. The exact immunogen used to generate this antibody is proprietary information. Database link P35613 |
| **Clone number** | MEM-M6/1 |
| **Purification technique** | Affinity purification Protein A |
| **Specificity** | Human CD147 antigen. This antibody recognizes an epitope in the N-terminal Ig domain (D1). This high-affinity antibody is capable of binding to unstimulated peripheral blood T cells. |
| **Light chain type** | kappa |
| **Concentration** | 1 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |
| **Purification notes** | Purified from TCS. Purity >95% by SDS-PAGE. |

## Reactivity data

## IP

### Expected

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|---|---|
| **Species** | Human |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

## Flow Cyt

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 10 µg/mL |
| **Notes** | ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody. |

## WB

### Tested

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|---|---|
| **Species** | Human |
| **Dilution info** | 1 µg/mL |
| **Notes** | Both reducing and non-reducing conditions. |

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 10 µg/mL |
| **Notes** | - |

# Target data

See full target information BSG

| | |
|---|---|
| **Function** | Isoform 1. Essential for normal retinal maturation and development (By similarity). Acts as a retinal cell surface receptor for NXNL1 and plays an important role in NXNL1-mediated survival of retinal cone photoreceptors (PubMed:25957687). In association with glucose transporter SLC16A1/GLUT1 and NXNL1, promotes retinal cone survival by enhancing aerobic glycolysis and accelerating the entry of glucose into photoreceptors (PubMed:25957687). May act as a potent stimulator of IL6 |

secretion in multiple cell lines that include monocytes (PubMed:21620857). Isoform 1. (Microbial infection) Erythrocyte receptor for P.falciparum RH5 which is essential for erythrocyte invasion by the merozoite stage of P.falciparum isolates 3D7 and Dd2. Isoform 2. Signaling receptor for cyclophilins, essential for PPIA/CYPA and PPIB/CYPB-dependent signaling related to chemotaxis and adhesion of immune cells (PubMed:11688976, PubMed:11943775). Plays an important role in targeting monocarboxylate transporters SLC16A1/GLUT1, SLC16A11 and SLC16A12 to the plasma membrane (PubMed:17127621, PubMed:21778275, PubMed:28666119). Acts as a coreceptor for vascular endothelial growth factor receptor 2 (KDR/VEGFR2) in endothelial cells enhancing its VEGFA-mediated activation and downstream signaling (PubMed:25825981). Promotes angiogenesis through EPAS1/HIF2A-mediated up-regulation of VEGFA (isoform VEGF-165 and VEGF-121) and KDR/VEGFR2 in endothelial cells (PubMed:19837976). Plays a key role in regulating tumor growth, invasion, metastasis and neoangiogenesis by stimulating the production and release of extracellular matrix metalloproteinases and KDR/VEGFR2 by both tumor cells and stromal cells (fibroblasts and endothelial cells) (PubMed:11992541, PubMed:12553375, PubMed:15833850). Isoform 2. (Microbial infection) Erythrocyte receptor for P.falciparum RH5 which is essential for erythrocyte invasion by the merozoite stage of P.falciparum isolates 3D7, Dd2, 7G8 and HB3 (PubMed:22080952, PubMed:26195724). Binding of P.falciparum RH5 results in BSG dimerization which triggers an increase in intracellular Ca(2+) in the erythrocyte (PubMed:28409866). This essential step leads to a rearrangement of the erythrocyte cytoskeleton required for the merozoite invasion (PubMed:28409866). Isoform 2. (Microbial infection) Can facilitate human SARS coronavirus (SARS-CoV-1) infection via its interaction with virus-associated PPIA/CYPA. Isoform 2. (Microbial infection) Can facilitate HIV-1 infection via its interaction with virus-associated PPIA/CYPA. Isoform 2. (Microbial infection) First described as a receptor for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), it is not required for SARS-CoV-2 infection. Isoform 2. (Microbial infection) Acts as a receptor for measles virus. Isoform 2. (Microbial infection) Promotes entry of pentamer-expressing human cytomegalovirus (HCMV) into epithelial and endothelial cells. Plays a role in neurite outgrowth.

## Storage

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|---|---|
| **Shipped at conditions** | Blue Ice |
| **Appropriate short-term storage conditions** | +4°C |
| **Appropriate long-term storage conditions** | +4°C |
| **Storage information** | Do Not Freeze |

## Notes

This product was changed from ascites to tissue culture supernatant on 24th January 2018. Please note that the dilutions may need to be adjusted accordingly. If you have any questions, please do not hesitate to contact our

scientific support team.

## Supplementary info

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

| | |
|---|---|
| **Activity summary** | CD147 also referred to as EMMPRIN or basigin is a transmembrane glycoprotein with a molecular weight of roughly 50–60 kDa. This protein is found on the surface of many cell types including leukocytes platelets and endothelial cells and it plays a critical role in cell function related to immune responses and cellular interactions. CD147 interacts with several cellular components serving mechanical functions such as facilitating cell-to-cell communication and contributing to the stability of cell structures. The protein is ubiquitously expressed but exhibits higher levels in tissues like the brain skin and liver. |
| **Biological function summary** | CD147 engages in multiple roles beyond its mechanical functions. It functions as a part of protein complexes and is involved in processes such as regulation of matrix metalloproteinases (MMPs) which are pivotal in tissue remodeling and repair. It can modulate inflammatory responses and is essential for cellular signaling pathways that affect cellular metabolism and growth. The involvement of CD147 in immune responses indicates its importance in various physiological processes. |
| **Pathways** | CD147 plays a significant role in the MAPK and NF-kB signaling pathways which are fundamental for controlling inflammatory responses and cell survival. It interacts with proteins such as integrins and cyclophilins which are important for mediating these pathways. These interactions highlight CD147's impact on cellular dynamics and its potential role in modulating the cellular environment in response to external stimuli. |
| **Associated diseases and disorders** | CD147 has been implicated in cancer promotion and progression as well as inflammatory diseases like rheumatoid arthritis. Its overexpression is often observed in tumors where it interacts with MMPs to facilitate tumor invasion and metastasis. In inflammatory conditions CD147 can influence the behavior of proteins like cytokines exacerbating symptoms and contributing to disease severity. These associations make CD147 a potential target for therapeutic intervention in cancer and inflammatory diseases. |

## 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.

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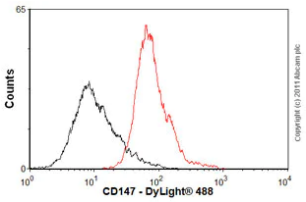
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In the unlikely event of one of our products not working as expected, you are covered by our product promise.

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## 6 product images

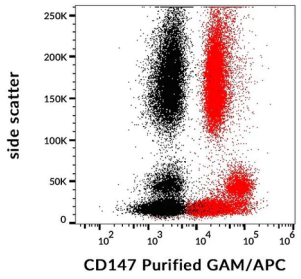



### Flow Cytometry - Anti-CD147 antibody [MEM-M6/1] (ab666)

CD147 Flow Cytometry staining using mouse Anti-CD147 antibody

Overlay histogram showing peripheral blood lymphocytes stained with ab666 (red line). The cells were incubated with the antibody (ab666, 1 µg/1x$10^6$ cells) for 30 minutes at 4°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 minutes at 4°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2 µg/1x$10^6$ cells) used under the same conditions. Acquisition of >5,000 events was performed gating on peripheral blood lymphocytes.

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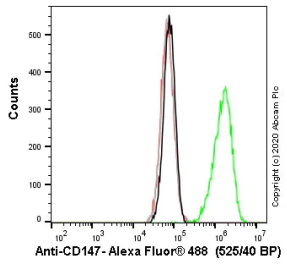



### Flow Cytometry - Anti-CD147 antibody [MEM-M6/1] (ab666)

CD147 Flow Cytometry staining using mouse Anti-CD147 antibody

Flow cytometry of human peripheral blood cells with ab666 at 1 µg/ml

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### Flow Cytometry - Anti-CD147 antibody [MEM-M6/1] (ab666)

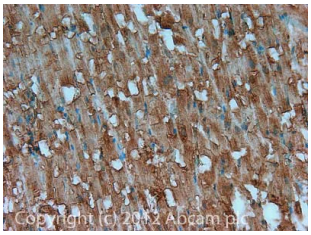
CD147 Flow Cytometry staining using mouse Anti-CD147 antibody

Flow cytometry overlay histogram showing wild-type A549 (green line) and BSG knockout A549 cells (ab273748) stained with ab666 (red line). The cells were incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab666) (1x$10^6$ in 100µl at 10 µg/ml) for 30 min at 4°C.

The secondary antibody Goat anti-mouse IgG H&L (Alexa Fluor® 488, pre-adsorbed) (ab150117) was used at 1/2000 for 30 min at 4°C.

Isotype control antibody was mouse IgG1κ (ab170190) used at the same concentration and conditions as the primary antibody (wild-type A549 - black line; BSG knockout A549 - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

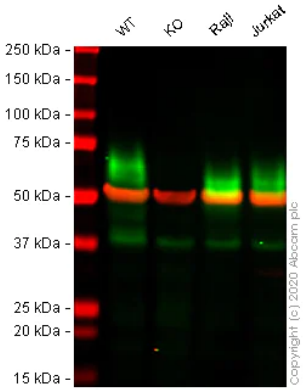
Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.




## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD147 antibody [MEM-M6/1] (ab666)

CD147 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-CD147 antibody

IHC image of CD147 staining in human normal heart muscle formalin fixed paraffin embedded tissue section, performed on a Leica BondTM system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6, epitope retrieval solution 1) for 20 minutes. The section was then incubated with ab666, 5 µg/ml, for 15 minutes at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.




## Western blot - Anti-CD147 antibody [MEM-M6/1] (ab666)

CD147 Western blot staining using mouse Anti-CD147 antibody

**Lanes 1 - 4:** Merged signal (red and green). Green - ab666 observed at 55-70 kDa. Red - loading control ab52866 (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55kDa.
ab666 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 ab666 and ab52866 (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4°C at 1 µg/ml and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (ab216777) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-CD147 antibody [MEM-M6/1] (ab666) at 1 µg/mL

Lane 1:
Wild-type A549 cell lysate at 30 µg

Lane 2:
Western blot - Human BSG (CD147) knockout A549 cell lysate (ab275500) at 20 µg

Lane 3:
Raji cell lysate at 30 µg

Lane 4:
Jurkat cell lysate at 30 µg

Performed under reducing conditions.

Predicted band size: 42 kDa

Observed band size: 55-70 kDa

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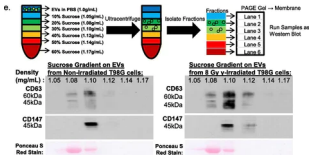


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## Western blot - Anti-CD147 antibody [MEM-M6/1] (ab666)

CD147 western blot using anti-CD147 antibody [MEM-M6/1] ab666. Publication image and figure legend from Colangelo, N. W. & Azzam, E. I., 2020, Cell Commun Signal, PubMed 32033611.

ab666 was used in this publication in western blot. This may not be the same as the application(s) guaranteed by Abcam. For a full list of applications guaranteed by Abcam for ab666 please see the product overview.

Irradiation of glioblastoma cells increases the levels of CD147 protein in their EVs. a Immunoblots for CD147 on the EVs of T98G and U-87 MG cells are shown for the 0 or 8 Gy dose at 24, 48, and 72 h for three experiments. The ‘C’ label represents the 0 Gy control samples, and the ‘IR’ label is for samples receiving 8 Gy of ionizing radiation. Immunoblot for CD147 in lysates of T98G cells, harvested after the 72 h collection of EVs, was also performed. CD147 levels were normalized relative to the Ponceau S stain. Graphical representations of these results are also displayed. b Immunoblot for CD147 on EVs collected at 24 h from T98G cells receiving increasing doses of γ-rays. The level of CD147 in the cells at 24 h was also measured. c Immunoblot for CD147 in EVs from T98G cells with CD147 knockdown, showing that CD147 still increases in response to γ-irradiation, even in partial knockdowns. Plasmid 1 clone 1 was the knockdown used in experiments. The scramble control (Scr) has saturated signal due to the high levels of CD147 in its EVs relative to the knockdowns. Clones 2 and 3 of plasmid 1 and clones 1, 3, and 4 of plasmid 2

demonstrate that even following knockdown, CD147 is increased in EVs when cells are irradiated. Immunoblots for CD147 on the cell lysates of the selected clones is also shown relative to a control c to provide a relative knockdown efficiency for cellular CD147. d Immunoblots on EVs of 0 or 8 Gy γ-irradiated U-118 MG cells, with the EVs from γ-irradiated U-87 MG cells used as a positive control. Despite EVs from U-118 MG cells not having CD147, they contained MCT1 and MCT4, which are found in EVs and co-localize with CD147. Ponceau S Red Stain was used as loading control. e Sucrose gradients determined that CD147 and CD63 sediment together at the correct density for EVs

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