abcam

ab308460

Anti-CD58 antibody [EPR27435-85] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR27435-85
Purification technique	Affinity purification Protein A
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

WB

Tested

Species	Human
Dilution info	-

Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

IHC-P

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information CD58

Function Ligand of the T-lymphocyte CD2 glycoprotein. This interaction is important in mediating thymocyte interactions with thymic epithelial cells, antigen-independent and -dependent interactions of T-lymphocytes with target cells and antigen-presenting cells and the T-lymphocyte rosetting with erythrocytes. In addition, the LFA-3/CD2 interaction may prime response by both the CD2+ and LFA-3+ cells.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C

Notes

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.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. 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.

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.

Compatibility
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.

Supplementary info

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

Activity summary	CD58 also known as lymphocyte function-associated antigen 3 (LFA-3) is a glycoprotein with a molecular mass of approximately 60 kDa. It is present on the surface of various cell types including antigen-presenting cells epithelial cells and some other leukocytes. This protein serves as a ligand for CD2 which is expressed on T cells and natural killer cells. Through its interaction with CD2 CD58 mediates cell adhesion and plays a role in immunological synapse formation which is essential for effective immune responses.
Biological function summary	Lymphocyte function-associated antigen 3 acts as a significant participant in the immune system. It plays a role in T cell activation and facilitates interactions between T cells and antigen-presenting cells. CD58 forms part of a complex network of cell surface proteins that enhances immune surveillance and response. Its role highlights its importance in adaptive immunity by promoting cell-to-cell communication and activation.
Pathways	CD58 participates in the cell adhesion pathways. It interacts closely with CD2 which is involved in the activation and signaling of T cells. Through this interaction CD58 plays roles in pathways like T cell receptor signaling and immune response. It enhances cell-cell adhesion helping T cells maintain their position during immune surveillance and activation.

Associated diseases and disorders	CD58 relates to conditions such as autoimmune diseases and certain cancers. In autoimmune disorders aberrant expression or function of CD58 may contribute to inappropriate immune activation leading to tissue damage. In cancer CD58 alterations can affect tumor immune evasion influencing how the immune system recognizes and attacks cancer cells. In multiple sclerosis an autoimmune disease the interaction between CD58 and CD2 has been studied providing insights into its role in disease mechanisms.

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

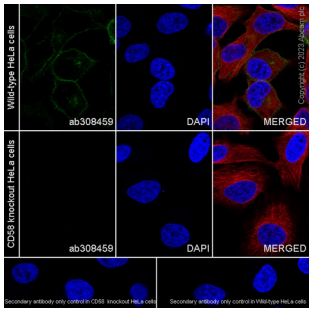
We do not recommend this combination. It is not covered by our product promise.

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.

6 product images

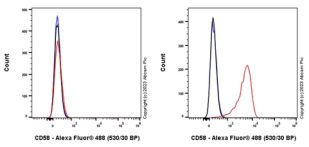


Immunocytochemistry/ Immunofluorescence - Anti-CD58 antibody [EPR27435-85] - BSA and Azide free (ab308460)

CD58 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CD58 antibody

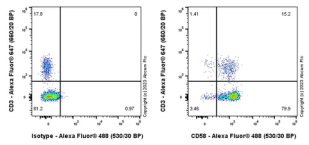
This data was developed using ab308459, the same antibody clone in a different buffer formulation. Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized CD58 KO HeLa (CD58 knockout human cervix adenocarcinoma epithelial cell) (ab265947); Parental HeLa (parental human cervix adenocarcinoma epithelial cell) cells labelling CD58 with ab308459 at 1/50 (10.08 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2 ug/ml dilution (Green). Confocal image

showing membranous staining in parental HeLa, and no staining CD58 KO HeLa cell line. Image was taken with a confocal microscope(Leica-Microsystems, TCS SP8). ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5ug/ml dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2 ug/ml dilution.



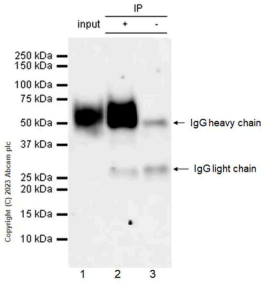
Flow Cytometry - Anti-CD58 antibody [EPR27435-85] - BSA and Azide free (ab308460)

This data was developed using ab308459, the same antibody clone in a different buffer formulation. Flow cytometric analysis of CD58 KO HeLa (ab265947) (human cervical adenocarcinoma epithelial cell, Left) / Parental HeLa (Right) cells labelling CD58 with ab308459 at 1/500 dilution (0.1 ug)/Red compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody. Gated on viable cells.



Flow Cytometry - Anti-CD58 antibody [EPR27435-85] - BSA and Azide free (ab308460)

This data was developed using ab308459, the same antibody clone in a different buffer formulation. Flow cytometric analysis of Human PBMC (human peripheral blood mononuclear cell) cells labelling CD58 with ab308459 at 1/500 dilution (0.1 ug)/Red compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody. Cells were co-stained with anti-CD3 conjugated to 647. Gated on viable cells.



Immunoprecipitation - Anti-CD58 antibody [EPR27435-85] - BSA and Azide free (ab308460)

This data was developed using ab308459, the same antibody clone in a different buffer formulation.
CD58 was immunoprecipitated from 0.35 mg Raji (human burkitt's lymphoma b lymphocyte) whole cell lysate 10 ug with ab308459 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab308459 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

Lane 1: Raji whole cell lysate

Lane 2: ab308459 IP in Raji whole cell lysate

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab308459 in Raji whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST

This blot was developed using a high sensitivity ECL substrate. The high-sensitivity ECL substrate used allows for the detection of proteins in the mid-femtogram range.

All lanes:
Immunoprecipitation - Anti-CD58 antibody [EPR27435-85] (ab308459) at 1/30 dilution

All lanes:
Raji (human burkitt's lymphoma b lymphocyte) whole cell lysate at 10 µg

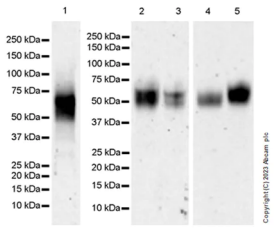
Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Developed using the ECL technique.

Observed band size: 55 kDa, 70 kDa

Exposure time: 102s



Western blot - Anti-CD58 antibody [EPR27435-85] - BSA and Azide free (ab308460)

This data was produced using ab308459, the same antibody clone but in a different formulation. Blocking and diluting buffer and concentration: 5% NFDM/TBST. The molecular weight observed is consistent with what has been described in the literature (PMID: 19268704; PMID: 3309127) and due to the high glycosylation of the protein. The blot of lane 1 was developed using a high sensitivity ECL substrate. Exposure time: 180 seconds.
All lanes:
Western blot - Anti-CD58 antibody [EPR27435-85] (ab308459) at 1/1000 dilution

Lane 1:
Human kidney tissue lysate at 20 µg

Lane 2:
Human tonsil tissue lysate at 20 µg

Lane 3:
Human placenta tissue lysate at 20 µg

Lane 4:
Jurkat (human t cell leukemia t lymphocyte from peripheral blood) whole cell lysate at 20 µg

Lane 5:
K-562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate at 20 µg

Secondary

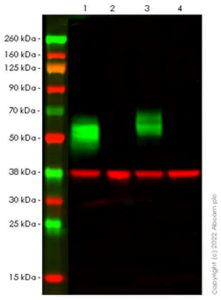
Lanes 1 - 5:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/2000 dilution

Lanes 1 - 5:
Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/20000 dilution

Predicted band size: 28 kDa

Observed band size: 55-70 kDa

Exposure time: 180s



Western blot - Anti-CD58 antibody [EPR27435-85] - BSA and Azide free (ab308460)

This data was produced using ab308459, the same antibody clone but in a different formulation. Blocking and diluting buffer and concentration: Intercept® (TBS) Blocking Buffer diluted with an equal volume of TBS. Lysates at 20 µg per lane. The samples were run on a Bis-Tris gel under reducing conditions. Western blot: Anti-CD58 antibody (ab308459) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20, 000 dilution, shown in red. In Western blot, ab308459 was shown to bind specifically to CD58. Target of interest was observed at 55-70 kDa in wild-type HeLa cell lysates (lane 1) with no signal observed at this size in CD58 knockout cell line ab265947 (knockout cell lysate ab257880)(lane 2). To generate this image, samples were first run on an SDS-PAGE gel then transferred onto an immobilon-FL PVDF membrane. Membranes were blocked in a fluorescent western blot (TBS-based) blocking solution before incubation with primary antibodies overnight at 4 °C. Blots were washed in TBS-T, incubated with secondary antibodies Goat anti-Rabbit IgG H&L 800CW (ab216773) and Goat anti-Mouse IgG H&L 680RD (ab216776) at 1/20000 dilution for 1 h at room temperature, washed again then imaged. Low expression: A-204 (PMID: 26039213).
All lanes:
Western blot - Anti-CD58 antibody [EPR27435-85] (ab308459) at 1/1000 dilution

Lane 1:
Wild-type HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
CD58 knockout HeLa whole cell lysate at 20 µg

Lane 3:
Raji (human burkitt's lymphoma b lymphocyte) whole cell lysate at 20 µg

Lane 4:
A-204 (human muscle rhabdomyosarcoma cell) whole cell lysate at 20 µg

Secondary

Lanes 1 - 4:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Lanes 1 - 4:
Western blot - Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) at 1/10000 dilution

Predicted band size: 28 kDa

Observed band size: 55-70 kDa

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