abcam

Product datasheet

Anti-CD58 antibody [TS2/9] ab171087

KO VALIDATED

6 Images

Overview

Product name	Anti-CD58 antibody [TS2/9]
Description	Mouse monoclonal [TS2/9] to CD58
Host species	Mouse
Tested applications	**Suitable for:** ICC/IF, Flow Cyt, WB
Species reactivity	**Reacts with:** Mouse, Human
Immunogen	Full length protein corresponding to Human CD58. Native protein Database link: **P19256**
General notes	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

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.
Storage buffer	Constituent: 99% PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	TS2/9
Isotype	IgG1

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab171087 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		1/10 - 1/100.
Flow Cyt		Use at an assay dependent concentration. **ab170190** - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.
WB		1/100 - 1/500. Predicted molecular weight: 24 kDa.

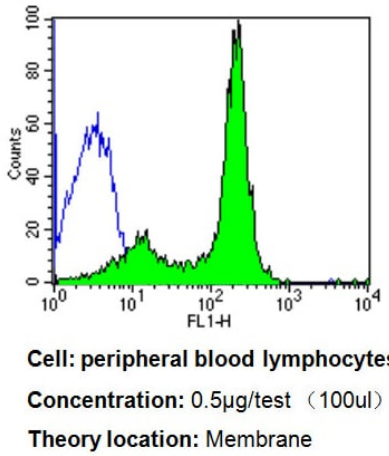
Target

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.

Sequence similarities Contains 1 Ig-like C2-type (immunoglobulin-like) domain.

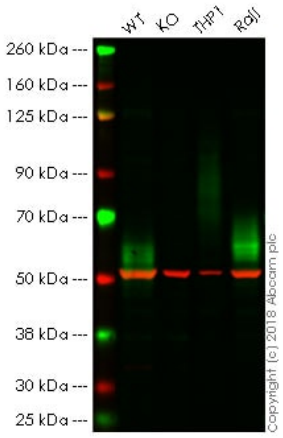
Cellular localization Cell membrane.

Images



Flow Cytometry - Anti-CD58 antibody [TS2/9] (ab171087)

Flow cytometry analysis of CD58 showing positive staining in the membrane of PBMC cells compared to an isotype control (blue). Human blood was collected, combined with a hydrophilic polysaccharide, centrifuged, transferred to a conical tube and washed with PBS. 50 ul of cell solution was added to each tube at a dilution of 2x10^7 cells/ml, followed by the addition of 50 ul of isotype control and ab171087 (0.5 ug/test). Cells were incubated for 30 min at 4ºC and washed with a cell buffer, followed by incubation with a DyLight 488-conjugated goat anti-mouse IgG (H+L) secondary for 30 min at 4ºC in the dark. FACS analysis was performed using 400 ul of cell buffer.



Western blot - Anti-CD58 antibody [TS2/9] (ab171087)

All lanes : Anti-CD58 antibody [TS2/9] (ab171087) at 1/250 dilution

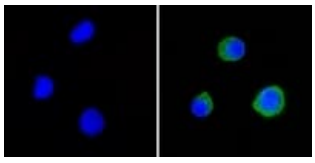
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : CD58 knockout HAP1 whole cell lysate
Lane 3 : THP1 whole cell lysate
Lane 4 : Raji whole cell lysate

Lysates/proteins at 40 µg per lane.

Predicted band size: 24 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab171087 observed at 43 kDa. Red - loading control, **ab176560**, observed at 50 kDa.

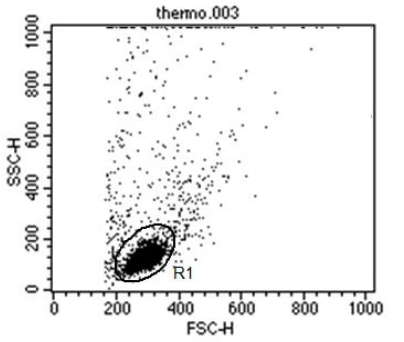
ab171087 was shown to specifically react with CD58 in wild-type HAP1 cells as signal was lost in CD58 knockout cells. Wild-type and CD58 knockout samples were subjected to SDS-PAGE. Ab171087 and **ab176560** (Rabbit anti alpha Tubulin loading control) were incubated overnight at 4°C at 1/250 dilution and 1/20000 dilution respectively. Blots were developed 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 1/20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-CD58 antibody [TS2/9] (ab171087)

Immunofluorescent analysis of CD58 (green) showing staining in the cytoplasm of Raji cells (right) compared to a negative control without primary antibody (left). Formalin-fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 5-10 minutes and blocked with 3% BSA-PBS for 30 minutes at room temperature. Cells were probed with a CD58 monoclonal antibody (ab171087) in 3% BSA-PBS at a dilution of 1:20 and incubated overnight at 4ºC in a humidified chamber. Cells were washed with PBST and incubated with a DyLight-conjugated secondary antibody in PBS at room temperature in the dark. F-actin (red) was stained with a

fluorescent red phalloidin and nuclei (blue) were stained with Hoechst or DAPI. Images were taken at a magnification of 60x.



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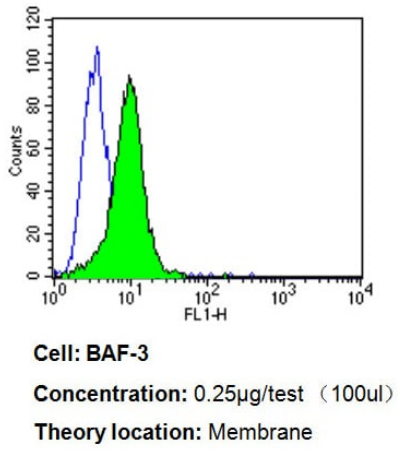
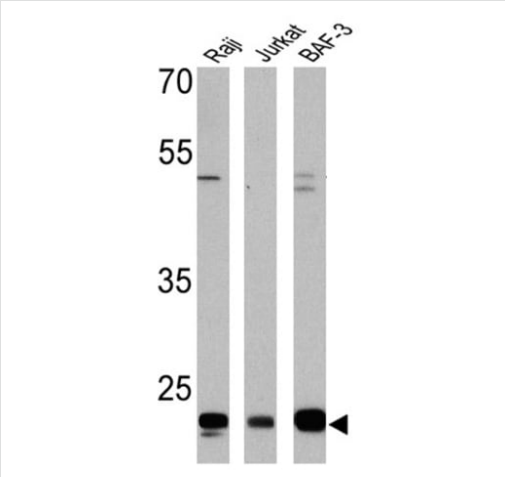


Flow Cytometry - Anti-CD58 antibody [TS2/9] (ab171087)

Flow cytometry analysis of CD58 showing weakly positive staining in the membrane of BAF-3 cells compared to an isotype control (blue). Cells were harvested, adjusted to a concentration of 1-5x10^6 cells/ml, fixed with 2% paraformaldehyde, washed with PBS, and incubated with ab171087 (0.25 ug/test) for 60 min at room temperature. Cells were then blocked in a solution of 2% BSA-PBS for 30 min at room temperature, incubated for 40 min at room temperature in the dark using a Dylight 488-conjugated goat anti-mouse IgG (H+L) secondary antibody, and re-suspended in PBS for FACS analysis.



Western blot - Anti-CD58 antibody [TS2/9] (ab171087)

All lanes : Anti-CD58 antibody [TS2/9] (ab171087) at 1/100 dilution

Lane 1 : Raji cell lysate
Lane 2 : Jurkat cell lysate
Lane 3 : BAF-3 cell lysate

Lysates/proteins at 25 µg per lane.

Predicted band size: 24 kDa
Observed band size: 24 kDa

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