abcam

Product datasheet

Anti-ICAM1 antibody [EPR24639-3] ab282575

敲除验证 重组 RabMAb

★★★★★ 1 Abreviews 4 References 13 图像

概述

产品名称	Anti-ICAM1抗体[EPR24639-3]
描述	兔单克隆抗体[EPR24639-3] to ICAM1
宿主	Rabbit
特异性	Rat species is recommended based on Flow Cyt result. We do not guarantee other applications for rat.
经测试应用	**适用于:** Flow Cyt, IP, IHC-P, ICC/IF, WB **不适用于:** IHC-Fr
种属反应性	**与反应:** Rat, Human
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: Wild-type HeLa whole cell lysate. Ramos and Raji whole cell lysate. Human kidney tissue lysate. IHC-P: Human kidney tissue. Human breast cancer tissue. ICC/IF: Raji cells. Wild-type HeLa cells. Flow Cyt: Rat splenocytes. C6 and Raji cells. IP: Raji whole cell lysate.
常规说明	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**.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59.94% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
纯度	Protein A purified

克隆	单克隆
克隆编号	EPR24639-3
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab282575于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt		1/500.
IP		1/30.
IHC-P		1/600. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/500.
WB	★★★★★ (1)	1/1000. Predicted molecular weight: 57 kDa.

应用说明 Is unsuitable for IHC-Fr.

靶标

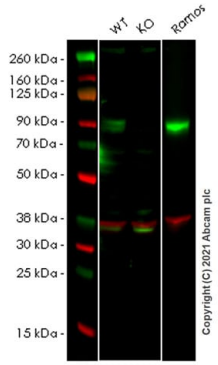
功能 ICAM proteins are ligands for the leukocyte adhesion protein LFA-1 (integrin alpha-L/beta-2). During leukocyte trans-endothelial migration, ICAM1 engagement promotes the assembly of endothelial apical cups through ARHGEF26/SGEF and RHOG activation. In case of rhinovirus infection acts as a cellular receptor for the virus.

序列相似性 Belongs to the immunoglobulin superfamily. ICAM family.
Contains 5 Ig-like C2-type (immunoglobulin-like) domains.

翻译后修饰 Monoubiquitinated, which is promoted by MARCH9 and leads to endocytosis.

细胞定位 Membrane.

图片



Western blot - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

All lanes : Anti-ICAM1 antibody [EPR24639-3] (ab282575) at 1/1000 dilution

Lane 1 : Wild-type HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 2 : ICAM1 knockout HeLa whole cell lysate
Lane 3 : Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (IRDye® 800CW) (**ab216773**) and Goat Anti-Mouse IgG H&L (IRDye® 680RD) (**ab216776**) at 1/10000 dilution

Predicted band size: 57 kDa
Observed band size: 90 kDa

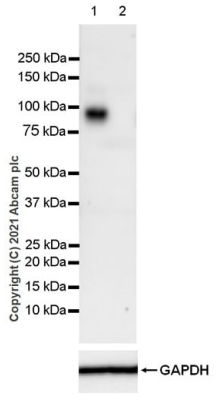
Blocking and diluting buffer and concentration: Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS

Lanes 1-3: Merged signal (red and green). Green - ab282575 observed at 90kDa. Red - loading control **ab8245** observed at 36 kDa.

ab282575 Anti-ICAM1 antibody [EPR24639-3] was shown to react with ICAM1 in wild-type Hela cells in Western blot. Loss of signal was observed when knockout cell line **ab261742** (knockout cell lysate **ab256947**) was used. Wild-type and ICAM1 knockout samples were subjected to SDS-PAGE.

ab282575 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated at 4°C overnight at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**)

and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

All lanes : Anti-ICAM1 antibody [EPR24639-3] (ab282575) at 1/1000 dilution

Lane 1 : Wild-type HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 2 : ICAM1 knockout Hela whole cell lysate

Lysates/proteins at 20 µg per lane.

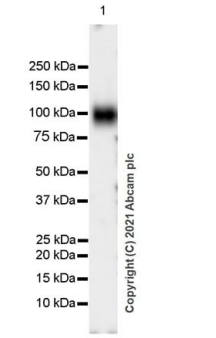
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 57 kDa
Observed band size: 90 kDa

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

ab282575 Anti-ICAM1 antibody [EPR24639-3] was shown to react with ICAM1 in wild-type Hela cells in Western blot. Loss of signal was observed when knockout cell line **ab261742** (knockout cell lysate **ab256947**) was used. Wild-type and ICAM1 knockout samples were subjected to SDS-PAGE.

Exposure time: 15 seconds.



Western blot - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

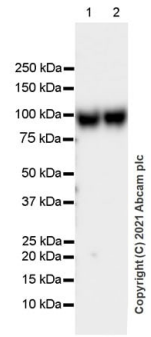
Anti-ICAM1 antibody [EPR24639-3] (ab282575) at 1/1000 dilution + human kidney tissue lysate at 20 µg

Secondary
VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at 1/1000 dilution

Predicted band size: 57 kDa
Observed band size: 90 kDa

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

Exposure time: 26 seconds



Western blot - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

All lanes : Anti-ICAM1 antibody [EPR24639-3] (ab282575) at 1/1000 dilution

Lane 1 : Ramos (human Burkitt's lymphoma B lymphocyte) whole cell lysate
Lane 2 : Raji (human Burkitts lymphoma B lymphocyte) whole cell lysate

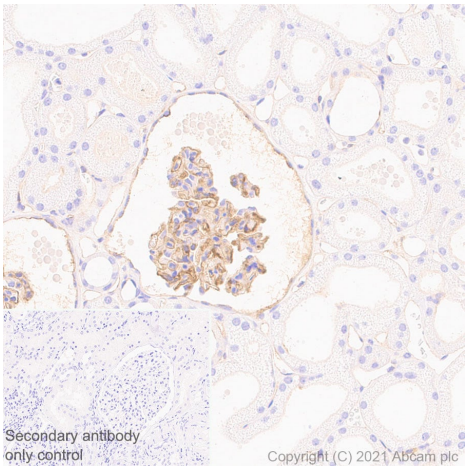
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 57 kDa
Observed band size: 90 kDa

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

Exposure time: 15 seconds

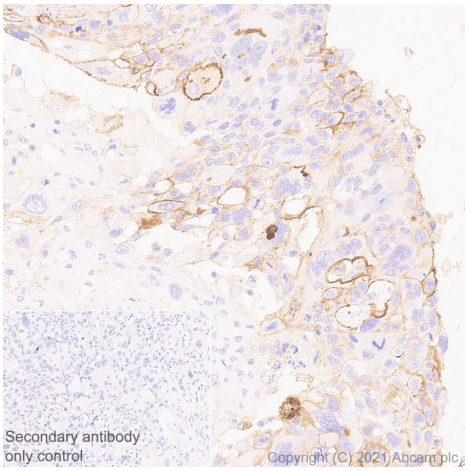


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

Immunohistochemical analysis of paraffin-embedded human kidney tissue labelling ICAM1 with ab282575 at 1/600 (0.902 µg/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on normal human glomerulus. The section was incubated with ab282575 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

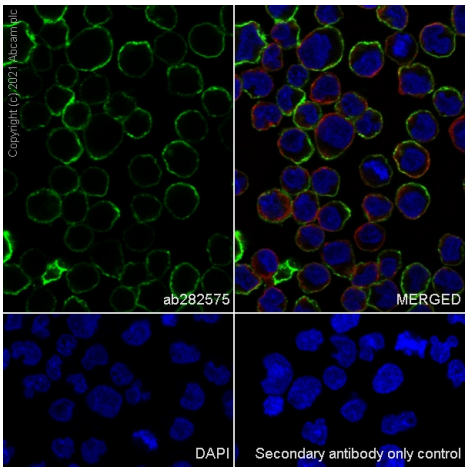


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

Immunohistochemical analysis of paraffin-embedded human breast cancer tissue labelling ICAM1 with ab282575 at 1/600 (0.902 µg/ml) dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on human breast cancer (PMID: 30082828). The section was incubated with ab282575 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

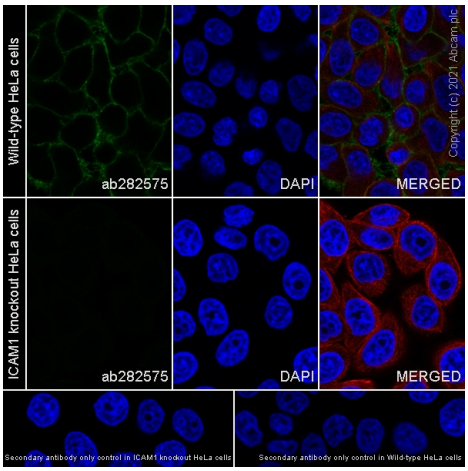


Immunocytochemistry/ Immunofluorescence - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Raji cells labelling ICAM1 with ab282575 at 1/500 (1.082 µg/ml) dilution, followed by **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing membranous staining in Raji cells.

ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.

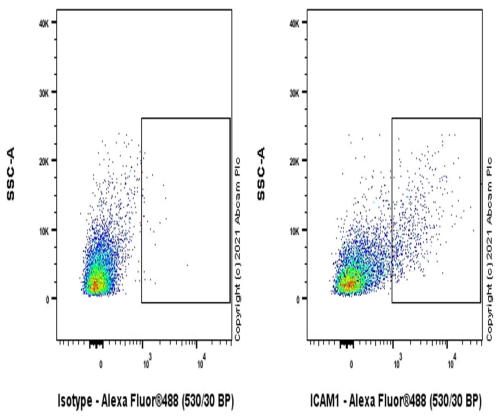


Immunocytochemistry/ Immunofluorescence - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized ICAM1 KO HeLa cells labelling ICAM1 with ab282575 at 1/500 (1.082 µg/ml) dilution, followed by **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution (Green). Confocal image showing membranous staining in wild-type HeLa cells, and no staining in ICAM1 knockout HeLa cells is observed.

ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The nuclear counterstain was DAPI (Blue).

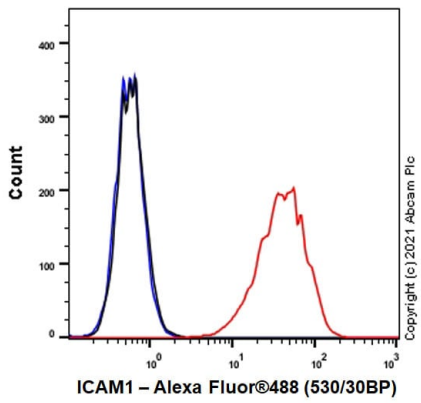
Secondary antibody only control: Secondary antibody is **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution.



Flow Cytometry - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

Flow cytometric analysis of rat splenocytes cells labelling ICAM1 with ab282575 at 1/500 dilution (0.1 µg)/ Right compared with a Rabbit monoclonal IgG (**ab172730**) / Left isotype control. A goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

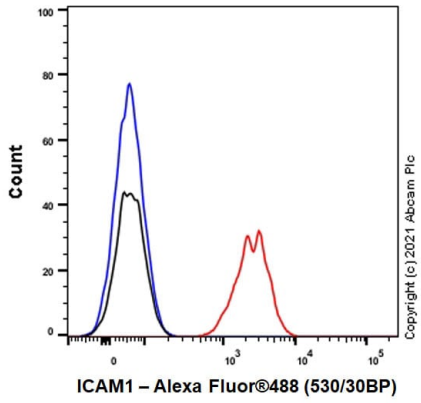
Gated on viable cells.



Flow Cytometry - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

Flow cytometric analysis of C6 (Rat glial tumor glial cell) cells labelling ICAM1 with ab282575 at 1/500 dilution (0.1 µg)/(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, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

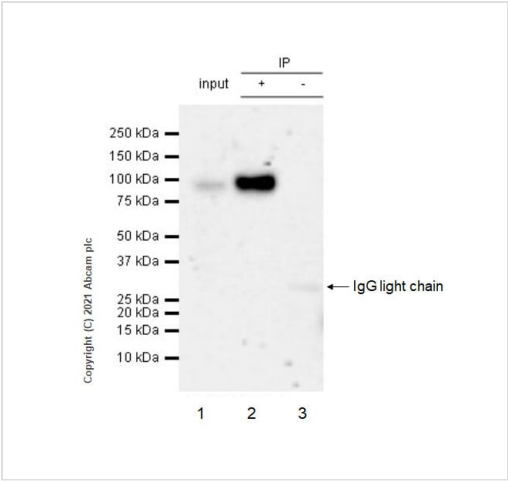
Gated on viable cells.



Flow Cytometry - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

Flow cytometric analysis of Raji (Human Burkitt's lymphoma B lymphocyte) cells labelling ICAM1 with ab282575 at 1/500 dilution (0.1 µg)/(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, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

Gated on viable cells.



Immunoprecipitation - Anti-ICAM1 antibody [EPR24639-3] (ab282575)

ICAM1 was immunoprecipitated from 0.35 mg Raji (human burkitt's lymphoma b lymphocyte) whole cell lysate 10 µg with ab282575 at 1/30 dilution (2 µg in 0.35 mg lysates). Western blot was performed on the immunoprecipitate using ab282575 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(**ab131366**) was used at 1/5000 dilution.

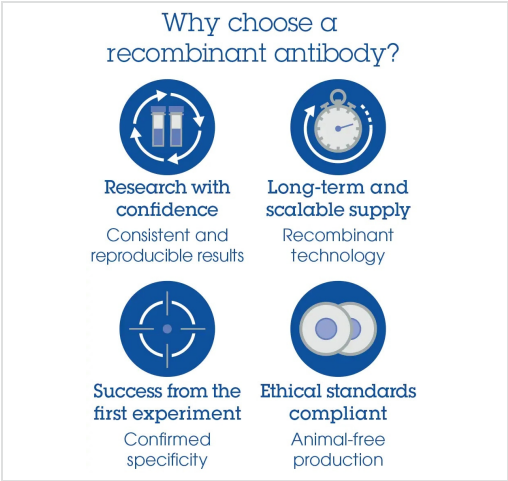
Lane 1: Raji whole cell lysate 10 µg

Lane 2: ab282575 IP in Raji whole cell lysate

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab282575 in Raji whole cell lysate

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

Exposure time: 3 minutes.



Anti-ICAM1 antibody [EPR24639-3] (ab282575)

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