abcam

Product datasheet

Anti-CD9 antibody [EPR23105-125] ab263019

KO VALIDATED Recombinant RabMAb

10 References 8 Images

Overview

Product name	Anti-CD9 antibody [EPR23105-125]
Description	Rabbit monoclonal [EPR23105-125] to CD9
Host species	Rabbit
Tested applications	**Suitable for:** IP, WB, IHC-P, Flow Cyt **Unsuitable for:** ICC/IF
Species reactivity	**Reacts with:** Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa, MCF7, HCT 116, HEL, Human tonsil and Human colon lysates. IHC-P: Human spleen and cervical carcinoma tissues. Flow Cyt: HCT 116 cells. IP: HCT 116 cell lysate.
General notes	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**.

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	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR23105-125

Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab263019 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

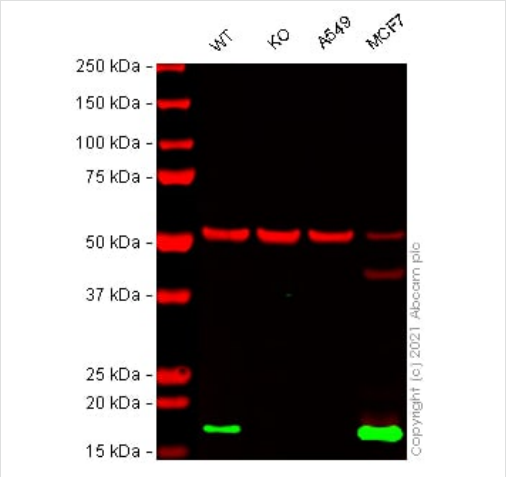
Application	Abreviews	Notes
IP		1/30.
WB		1/1000. Detects a band of approximately 22 kDa (predicted molecular weight: 25 kDa).
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
Flow Cyt		1/500.

Application notes Is unsuitable for ICC/IF.

Target

Function	Involved in platelet activation and aggregation. Regulates paranodal junction formation. Involved in cell adhesion, cell motility and tumor metastasis. Required for sperm-egg fusion.
Tissue specificity	Expressed by a variety of hematopoietic and epithelial cells.
Sequence similarities	Belongs to the tetraspanin (TM4SF) family.
Post-translational modifications	Protein exists in three forms with molecular masses between 22 and 27 kDa, and is known to carry covalently linked fatty acids.
Cellular localization	Membrane.

Images



Western blot - Anti-CD9 antibody [EPR23105-125] (ab263019)

All lanes : Anti-CD9 antibody [EPR23105-125] (ab263019) at 1/1000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD9 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

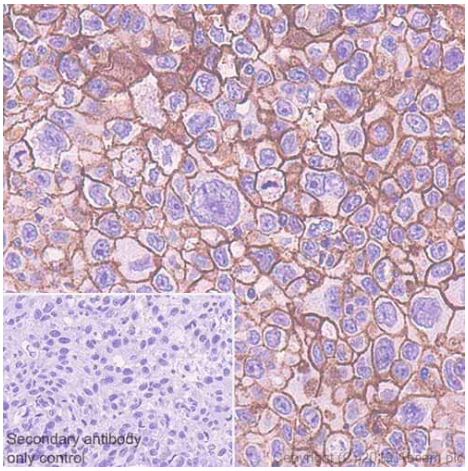
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 25 kDa
Observed band size: 18 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab263019 observed at 18 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

ab263019 was shown to react with CD9 in wild-type HeLa cells in Western blot with loss of signal observed in CD9 knockout cell line **ab255375** (CD9 knockout cell lysate **ab263754**). Wild-type HeLa and CD9 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with ab263019 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 h at room temperature before imaging.

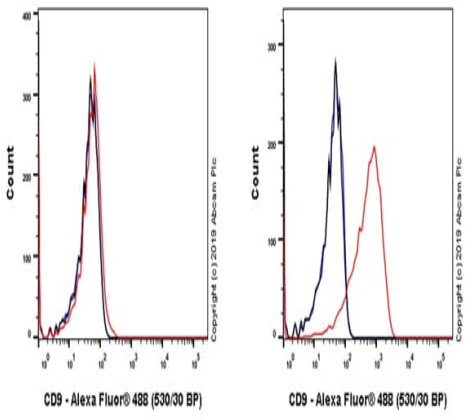


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD9 antibody [EPR23105-125] (ab263019)

Immunohistochemical analysis of paraffin-embedded Human cervical carcinoma tissue labeling CD9 with ab263019 at 1/1000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Positive staining on human cervical carcinoma. The section was incubated with ab263019 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 Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20mins.

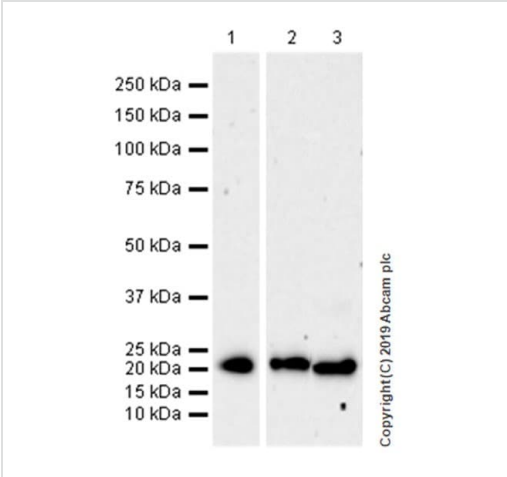


Flow Cytometry - Anti-CD9 antibody [EPR23105-125] (ab263019)

Flow cytometric analysis of Raji (human Burkitt's lymphoma B lymphocyte, Left) / HCT 116 (human colorectal carcinoma epithelial cell, Right) cells labelling CD9 with ab263019 at 1/500 dilution 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 was used as the secondary antibody.

Negative control: Raji (PMID: 8921952).

Gated on viable cells.



Western blot - Anti-CD9 antibody [EPR23105-125] (ab263019)

All lanes : Anti-CD9 antibody [EPR23105-125] (ab263019) at 1/1000 dilution

Lane 1 : HEL (erythroleukemia erythroblast), whole cell lysate
Lane 2 : Human tonsil lysate
Lane 3 : Human colon lysate

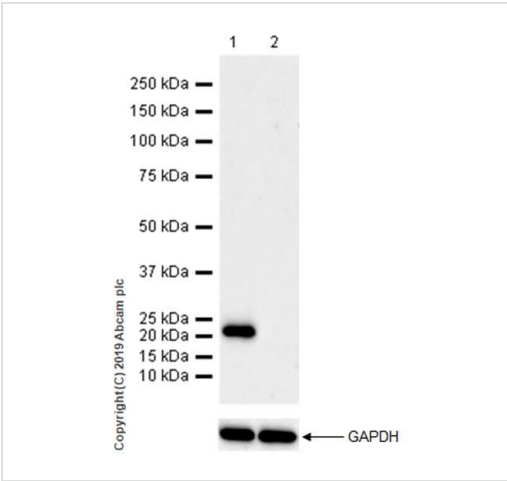
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 25 kDa
Observed band size: 22 kDa

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

The molecular weight observed is consistent with what has been described in the literature (PMID: 3275469). Exposure time: 3 minutes.



Western blot - Anti-CD9 antibody [EPR23105-125] (ab263019)

All lanes : Anti-CD9 antibody [EPR23105-125] (ab263019) at 1/1000 dilution

Lane 1 : HCT 116 (human colorectal carcinoma epithelial cell) 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 1/100000 dilution

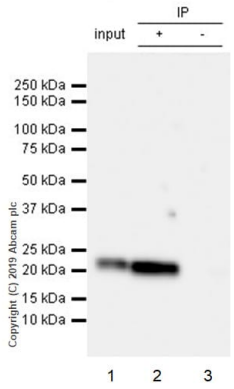
Predicted band size: 25 kDa

Observed band size: 22 kDa

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

The molecular weight observed is consistent with what has been described in the literature (PMID: 3275469).

Negative control: Raji (PMID: 8921952). Exposure time: 3 minutes.



Immunoprecipitation - Anti-CD9 antibody [EPR23105-125] (ab263019)

CD9 was immunoprecipitated from 0.35 mg HCT 116 (human colorectal carcinoma epithelial cell) whole cell lysate with ab263019 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab263019 at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/1000 dilution.

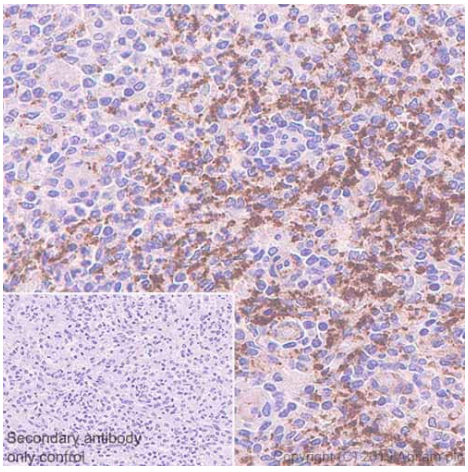
Lane 1: HCT 116 (human colorectal carcinoma epithelial cell) whole cell lysate 10ug.

Lane 2: ab263019 IP in HCT 116 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab263019 in HCT 116 whole cell lysate.

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

Exposure time: 10 seconds.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD9 antibody [EPR23105-125] (ab263019)

Immunohistochemical analysis of paraffin-embedded Human spleen tissue labeling CD9 with ab263019 at 1/1000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Positive staining on platelets of human spleen. The section was incubated with ab263019 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 Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

Anti-CD9 antibody [EPR23105-125] (ab263019)

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