

Product datasheet

Anti-EpCAM antibody [RM1016] α b282457

Recombinant **RabMAb**

[1 References](#) [17 Images](#)

Overview

Product name	Anti-EpCAM antibody [RM1016]
Description	Rabbit recombinant multiclonal [RM1016] to EpCAM
Host species	Rabbit
Tested applications	Suitable for: Flow Cyt, IP, WB, IHC-P, IHC-Fr, ICC
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	<p>This product was produced with the following immunogens:</p> <p>Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.</p> <p>Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.</p>
Positive control	<p>WB: HCT 116, 4T1 and NIH/3T3 whole cell lysate. Human colon lysate. Mouse and rat colon and kidney lysate. IHC-P: Human colon and colon carcinoma tissue. Mouse and rat colon tissue. Mouse colon carcinoma tissue. Human liver tissue. IHC-Fr: Mouse and rat colon tissue. ICC: HCT 116 and 4T1 cells. Flow Cyt: HCT-116 and 4T1 cells. IP: Human colon lysate. 4T1 whole cell lysate.</p>
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

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. Avoid freeze / thaw cycle.
Storage buffer	<p>pH: 7.2</p> <p>Preservative: 0.01% Sodium azide</p> <p>Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA</p>

Purity	Protein A purified
Clonality	Recombinant Multiclonal
Clone number	RM1016
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab282457 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
Flow Cyt		1/50.
IP		1/30.
WB		1/1000. Predicted molecular weight: 35 kDa.
IHC-P		1/4000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IHC-Fr		1/100. Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20)
ICC		1/250.

Target

Function	May act as a physical homophilic interaction molecule between intestinal epithelial cells (IECs) and intraepithelial lymphocytes (IELs) at the mucosal epithelium for providing immunological barrier as a first line of defense against mucosal infection. Plays a role in embryonic stem cells proliferation and differentiation. Up-regulates the expression of FABP5, MYC and cyclins A and E.
Tissue specificity	Highly and selectively expressed by undifferentiated rather than differentiated embryonic stem cells (ESC). Levels rapidly diminish as soon as ESC's differentiate (at protein levels). Expressed in almost all epithelial cell membranes but not on mesodermal or neural cell membranes. Found on the surface of adenocarcinoma.
Involvement in disease	Defects in EPCAM are the cause of diarrhea type 5 (DIAR5) [MIM:613217]. It is an intractable diarrhea of infancy characterized by villous atrophy and absence of inflammation, with intestinal epithelial cell dysplasia manifesting as focal epithelial tufts in the duodenum and jejunum. Defects in EPCAM are a cause of hereditary non-polyposis colorectal cancer type 8 (HNPCC8) [MIM:613244]. HNPCC is a disease associated with marked increase in cancer susceptibility. It is characterized by a familial predisposition to early-onset colorectal carcinoma (CRC) and extra-colonic tumors of the gastrointestinal, urological and female reproductive tracts. HNPCC is reported to be the most common form of inherited colorectal cancer in the Western world. Clinically, HNPCC is often divided into two subgroups. Type I is characterized by hereditary predisposition to colorectal cancer, a young age of onset, and carcinoma observed in the proximal colon. Type II is characterized by increased risk for cancers in certain tissues such as the

uterus, ovary, breast, stomach, small intestine, skin, and larynx in addition to the colon. Diagnosis of classical HNPCC is based on the Amsterdam criteria: 3 or more relatives affected by colorectal cancer, one a first degree relative of the other two; 2 or more generation affected; 1 or more colorectal cancers presenting before 50 years of age; exclusion of hereditary polyposis syndromes. The term 'suspected HNPCC' or 'incomplete HNPCC' can be used to describe families who do not or only partially fulfill the Amsterdam criteria, but in whom a genetic basis for colon cancer is strongly suspected. Note=HNPCC8 results from heterozygous deletion of 3-prime exons of EPCAM and intergenic regions directly upstream of MSH2, resulting in transcriptional read-through and epigenetic silencing of MSH2 in tissues expressing EPCAM.

Sequence similarities

Belongs to the EPCAM family.
Contains 1 thyroglobulin type-1 domain.

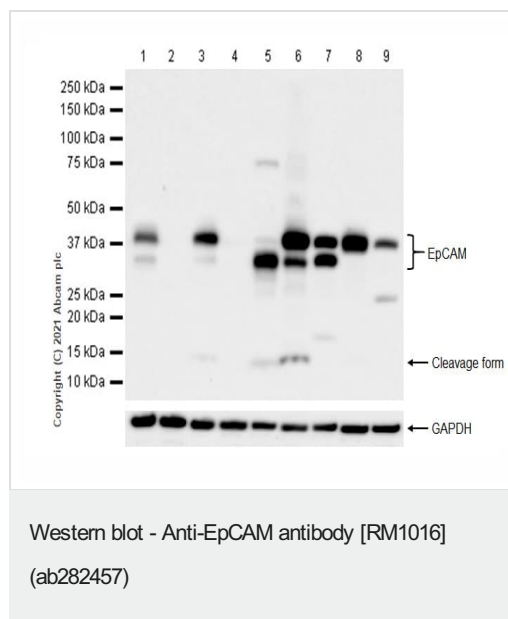
Post-translational modifications

Hyperglycosylated in carcinoma tissue as compared with autologous normal epithelia.
Glycosylation at Asn-198 is crucial for protein stability.

Cellular localization

Lateral cell membrane. Cell junction > tight junction. Co-localizes with CLDN7 at the lateral cell membrane and tight junction.

Images



All lanes : Anti-EpCAM antibody [RM1016] (ab282457) at 1/1000 dilution

Lane 1 : HCT 116 (Human colorectal carcinoma epithelial cell) whole cell lysate

Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 3 : 4T1 (Mouse mammary gland carcinoma epithelial cell) whole cell lysate

Lane 4 : NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate

Lane 5 : Human colon lysate

Lane 6 : Mouse colon lysate

Lane 7 : Rat colon lysate

Lane 8 : Mouse kidney lysate

Lane 9 : Rat kidney 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: 35 kDa

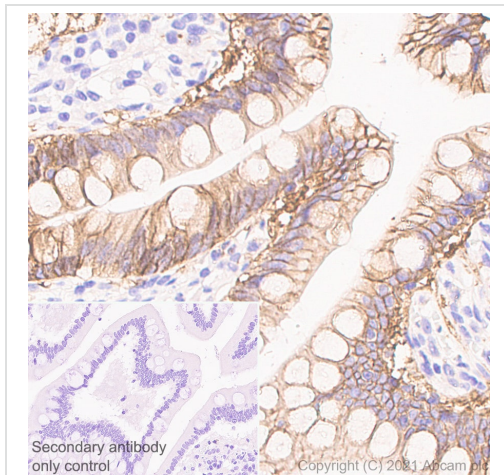
Observed band size: 12, 36, 40 kDa

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

Negative control: HeLa (PMID: 31806375), NIH/3T3 (PMID: 23264216).

The MW observed is consistent with the literature: PMID 19136966; PMID 23618806. The band between 12-15kDa has been documented in literature as the cleaved form of EpCAM (PMID: 23409978).

Exposure time: 8 seconds.

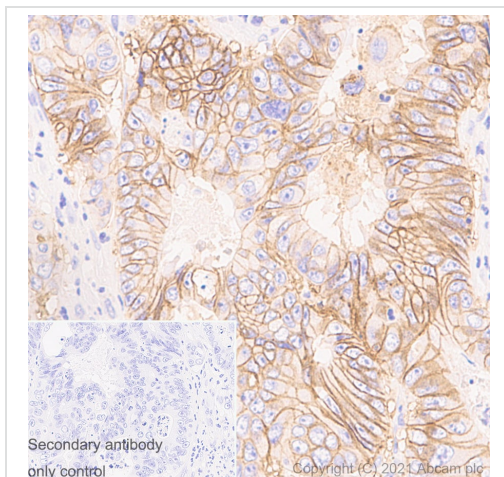


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of paraffin-embedded human colon tissue labeling EpCAM with ab282457 at 1/4000 (0.124 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Membranous staining on human colon. The section was incubated with ab282457 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.

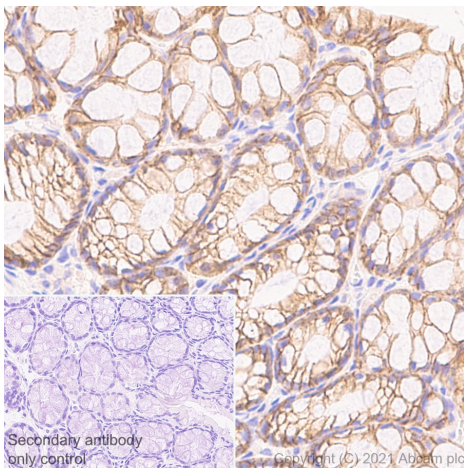


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue labeling EpCAM with ab282457 at 1/4000 (0.124 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Membranous staining on human colon carcinoma. The section was incubated with ab282457 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.

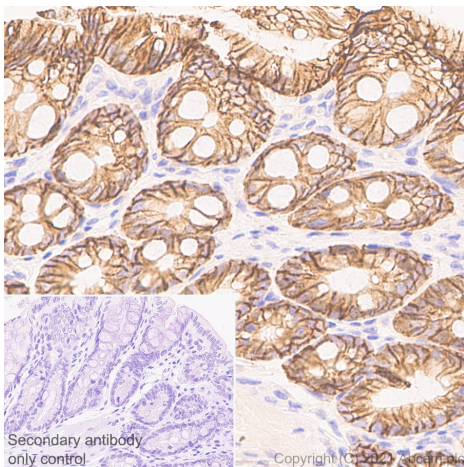


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of paraffin-embedded mouse colon tissue labeling EpCAM with ab282457 at 1/4000 (0.124 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Membranous staining on mouse colon. The section was incubated with ab282457 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.

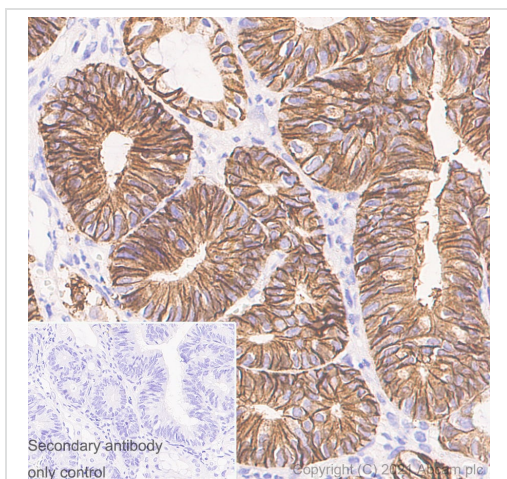


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of paraffin-embedded rat colon tissue labeling EpCAM with ab282457 at 1/4000 (0.124 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Membranous staining on rat colon. The section was incubated with ab282457 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.

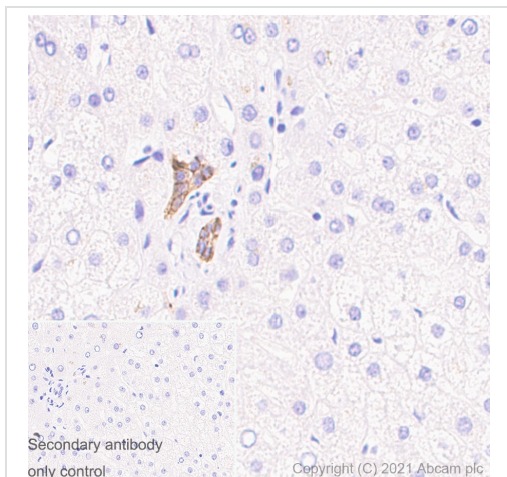


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of paraffin-embedded mouse colon carcinoma tissue labeling EpCAM with ab282457 at 1/4000 (0.124 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Membranous staining on mouse colon carcinoma. The section was incubated with ab282457 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.

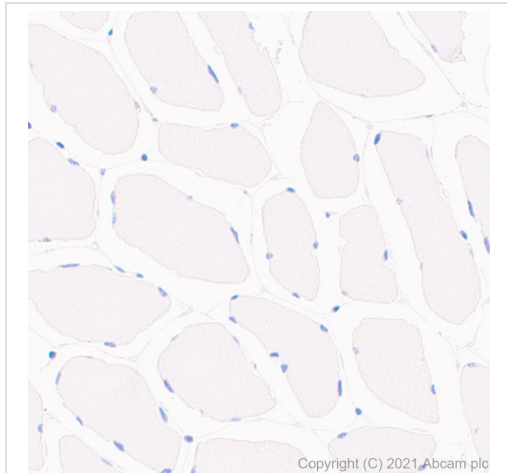


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of paraffin-embedded human liver tissue labeling EpCAM with ab282457 at 1/4000 (0.124 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Membranous staining on the bile duct in human liver, and no staining on the hepatocytes. The section was incubated with ab282457 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.

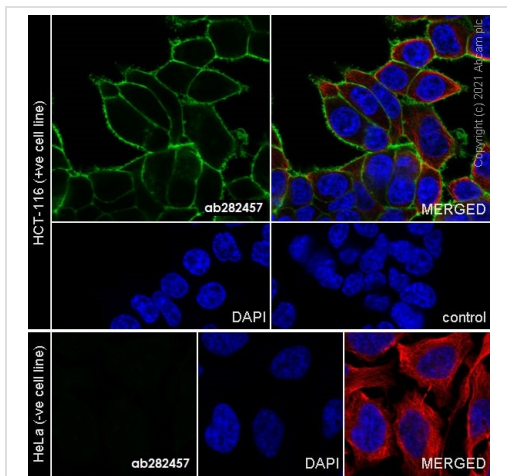


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of paraffin-embedded human skeletal muscle tissue labeling EpCAM with ab282457 at 1/4000 (0.124 µg/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). **Negative control:** no staining on human skeletal muscle. The section was incubated with ab282457 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.



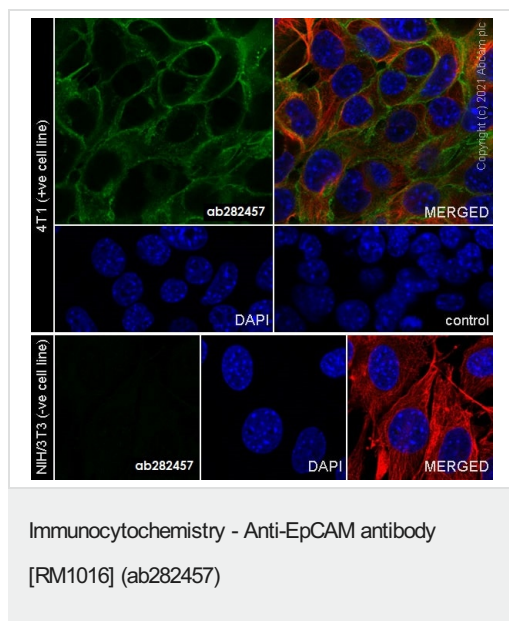
Immunocytochemistry - Anti-EpCAM antibody [RM1016] (ab282457)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HCT 116 and HeLa cells labelling EpCAM with ab282457 at 1/250 (1.976 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 HCT 116 cell line.

Negative control: HeLa (PMID: 31806375).

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.

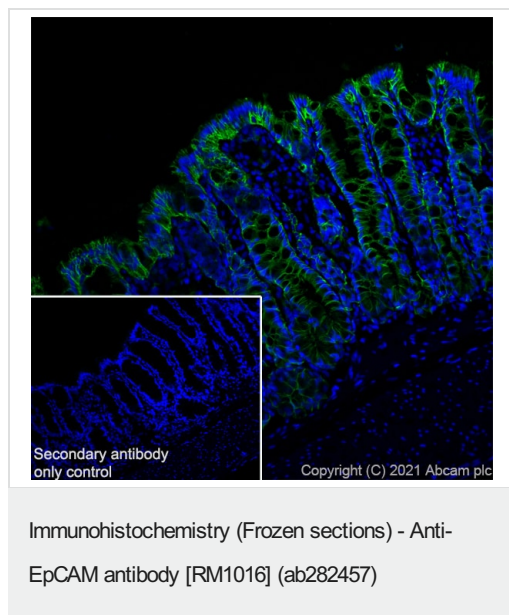


Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized 4T1 and NIH/3T3 cells labelling EpCAM with ab282457 at 1/250 (1.976 µg/ml) dilution, followed by **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution (Green). Confocal image showing membranous staining in 4T1 cell line.

Negative control: NIH/3T3 (PMID:31806375).

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).

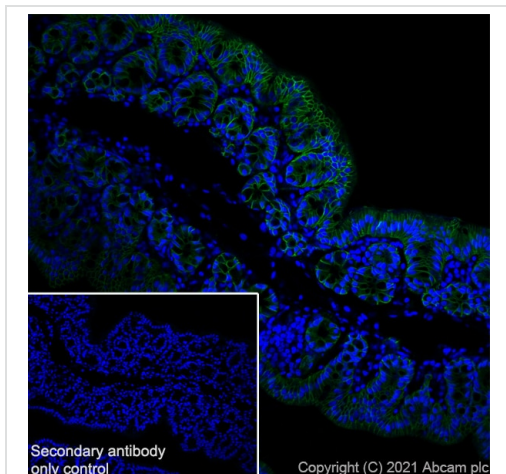
Secondary antibody only control: Secondary antibody is **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution.



Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen rat colon tissue labeling EpCAM with ab282457 at 1/100 (4.94 µg/ml) dilution followed by **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution (Green). Membranous staining on rat colon is observed. The nuclear counterstain was DAPI (Blue).

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

Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).

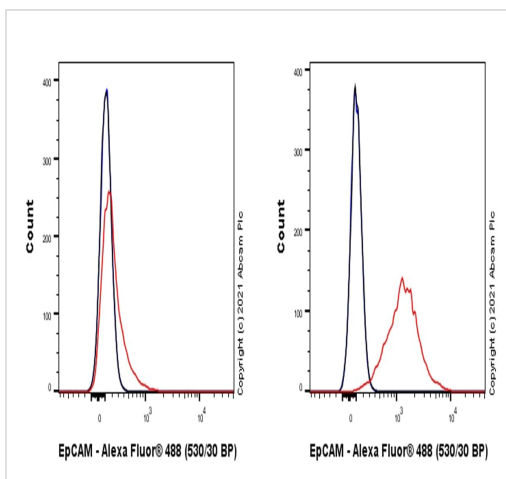


Immunohistochemistry (Frozen sections) - Anti-EpCAM antibody [RM1016] (ab282457)

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen mouse colon tissue labeling EpCAM with ab282457 at 1/100 (4.94 µg/ml) dilution followed by **ab150077** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution (Green). Membranous staining on mouse colon is observed. The nuclear counterstain was DAPI (Blue).

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

Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).

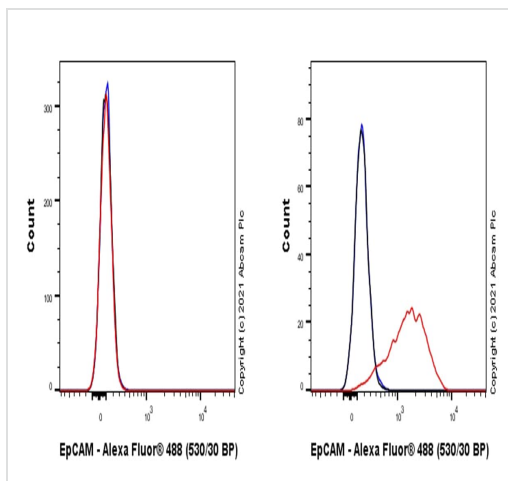


Flow Cytometry - Anti-EpCAM antibody [RM1016] (ab282457)

Flow cytometric analysis of HeLa (Human cervix adenocarcinoma epithelial cell, Left) / HCT-116 (Human colorectal carcinoma epithelial cell, Right) cells labelling EpCAM with ab282457 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.

Negative control: HeLa. (PMID: 31806375).

Gated on viable cells.

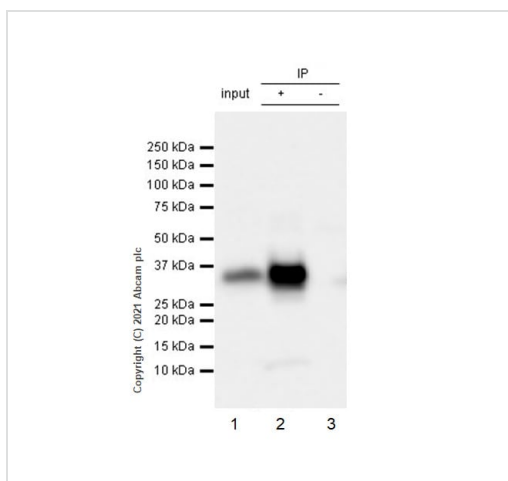


Flow Cytometry - Anti-EpCAM antibody [RM1016]
(ab282457)

Flow cytometric analysis of NIH/3T3 (Mouse embryonic fibroblast, Left) / 4T1 (Mouse mammary gland carcinoma epithelial cell, Right) cells labelling EpCAM with ab282457 at 1/50 dilution (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.

Negative control: NIH/3T3 (PMID:23264216).

Gated on viable cells.



Immunoprecipitation - Anti-EpCAM antibody
[RM1016] (ab282457)

EpCAM was immunoprecipitated from 0.35 mg human colon lysate 10 µg with ab282457 at 1/30 dilution (2 µg in 0.35 mg lysates).

Western blot was performed on the immunoprecipitate using ab282457 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(**ab131366**) was used at 1/5000 dilution.

Lane 1: Human colon lysate 10 µg

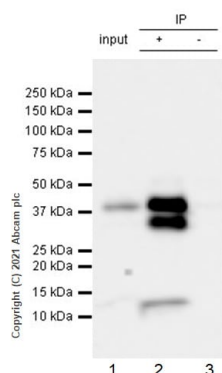
Lane 2: ab282457 IP in human colon lysate

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab282457 in human colon lysate

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

Exposure time: 8 seconds.

The MW observed is consistent with the literature: PMID 19136966; PMID 23618806. The band between 12-15kDa has been documented in literature as the cleaved form of EpCAM (PMID: 23409978).



Immunoprecipitation - Anti-EpCAM antibody
[RM1016] (ab282457)

EpCAM was immunoprecipitated from 0.35 mg 4T1 (Mouse mammary gland carcinoma epithelial cell) whole cell lysate 10 µg with ab282457 at 1/30 dilution (2 µg in 0.35 mg lysates). Western blot was performed on the immunoprecipitate using ab282457 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) ([ab131366](#)) was used at 1/5000 dilution.

Lane 1: 4T1 whole cell lysate 10 µg

Lane 2: ab282457 IP in 4T1 whole cell lysate

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab282457 in 4T1 whole cell lysate

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

Exposure time: 8 seconds.

The MW observed is consistent with the literature: PMID 19136966; PMID 23618806. The band between 12-15kDa has been documented in literature as the cleaved form of EpCAM (PMID: 23409978).

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-EpCAM antibody [RM1016] (ab282457)

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