abcam

Product datasheet

Anti-EpCAM antibody [EPR20532-225] ab223582

KO VALIDATED Recombinant RabMAb

★★★★★ 2 Abreviews 19 References 20 Images

Overview

Product name	Anti-EpCAM antibody [EPR20532-225]
Description	Rabbit monoclonal [EPR20532-225] to EpCAM
Host species	Rabbit
Tested applications	**Suitable for:** WB, IHC-P, Flow Cyt, IP, mIHC, ICC/IF
Species reactivity	**Reacts with:** Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Wild-type HAP1, HCT 116, and HT-29 cell lysates; Human breast cancer and fetal kidney lysates. IHC-P: Human colon and thyroid cancer tissues. ICC/IF: A431, T47D, HCT 116 and HT-29 cells. Flow Cyt: A431 and HCT 116 cells. IP: HCT 116 cell lysate. mIHC: Human breast cancer and endometrium tissues.
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: 0.05% BSA, 40% Glycerol, PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR20532-225

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab223582 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
WB		1/1000. Detects a band of approximately 42 kDa (predicted molecular weight: 35 kDa).
IHC-P	★★★★★ (1)	1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
Flow Cyt		1/500.
IP		1/30.
mIHC		1/500.
ICC/IF	★★★★★ (1)	1/500 - 1/5000.

Target

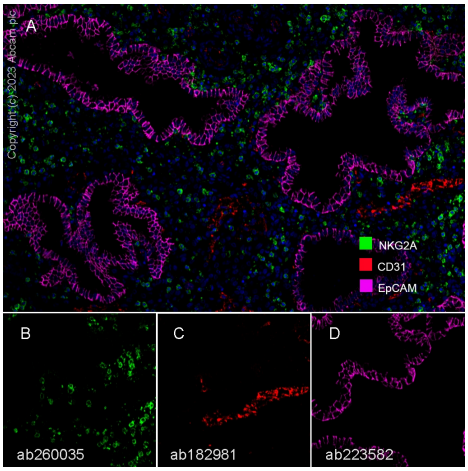
Function May act as a physical homophilic interaction molecule between intestinal epithelial cells (IECs) and intraepithelial lymphocytes (IELs) at the mucosal epithelium for providing immunological barrier as a first line of defense against mucosal infection. Plays a role in embryonic stem cells proliferation 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



Multiplex immunohistochemistry - Anti-EpCAM antibody [EPR20532-225] (ab223582)

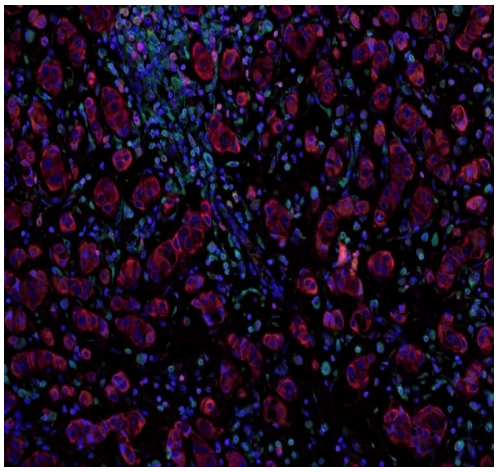
Fluorescence multiplex immunohistochemical analysis of the human endometrium (Formalin/PFA-fixed paraffin-embedded sections).

Panel A: merged staining of anti-EpCAM (ab223582, magenta; Opal™690), anti-NKG2A (**ab260035**, green; Opal™520) and anti-CD31 (**ab182981**, red; Opal™570) on human endometrium. Panel B: anti-NKG2A stained on NK cells. Panel C: anti-CD31 stained on endothelial cells. Panel D: anti-EpCAM stained on glandular cells. Opal Polymer HRP Ms + Rb was used as a secondary antibody.

The section was incubated in three rounds of staining: in the order of ab223582 at 1/500 dilution (1.008 µg/ml), **ab260035** at 1/2000 dilution (0.262 µg/ml) and **ab182981** at 1/4000 dilution (0.137 µg/ml) for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins. DAPI (blue) was used as a nuclear counter stain.



Multiplex immunohistochemistry - Anti-EpCAM antibody [EPR20532-225] (ab223582)

This image is courtesy of ImmunoAtlas.

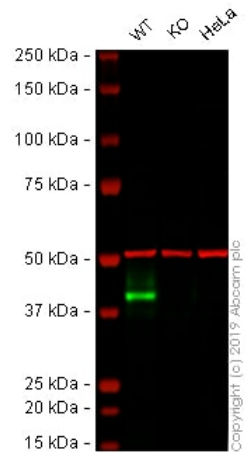
Fluorescence multiplex immunohistochemical analysis of Human breast cancer tissue (formalin-fixed paraffin-embedded section).

Merged staining of Anti-PD-L1 (**ab251611**; cyan; Opal™ 520), Anti-Granzyme B (**ab219803**; yellow; Opal™ 540), Anti-PD1 (**ab251613**; magenta; Opal™ 570), Anti-pan Cytokeratin (**ab264485**; red; Opal™ 620), Anti-EpCAM (**ab225894**; red; Opal™ 620), Anti-CD8 alpha (**ab251596**; green; Opal™ 650) and Anti-FOXP3 (**ab96048**; orange; Opal™ 690). EpCAM and pan-cytokeratin share the same dye and color. Dyes are pseudo-colored for better contrast of the markers.

The immunostaining was performed on a Leica Biosystems BOND® MAX instrument with an Opal™ 6-Plex Detection Kit (NEL821001KT, Akoya Biosciences®).

The section was incubated in six rounds of staining; sequentially for **ab251611** (1/750 dilution), **ab219803** (1/250 dilution), **ab251613** (1/750 dilution), **ab264485** (0.5 µg/ml), **ab225894** (1/1250 dilution), **ab251596** (1/1500 dilution) and **ab96048** (10 µg/ml); each using a separate fluorescent tyramide signal amplification system. EDTA based antigen retrieval (Leica Biosystems BOND® Epitope Retrieval Solution 2, pH 9.0, 20 minutes) was used in between rounds of tyramide signal amplification to remove the antibody from the previous round, to avoid any cross-reactivity. DAPI (dark blue) was used as a nuclear counter stain.
Microscopy and pseudocoloring of individual Opal™ dyes was performed using a Vectra 3 Imaging System (Akoya Biosciences®).

This data is courtesy of ImmunoAtlas and it can be found **here**.



Western blot - Anti-EpCAM antibody [EPR20532-225] (ab223582)

All lanes : Anti-EpCAM antibody [EPR20532-225] (ab223582) at 1/1000 dilution

Lane 1 : Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate
Lane 2 : EPCAM knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate
Lane 3 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

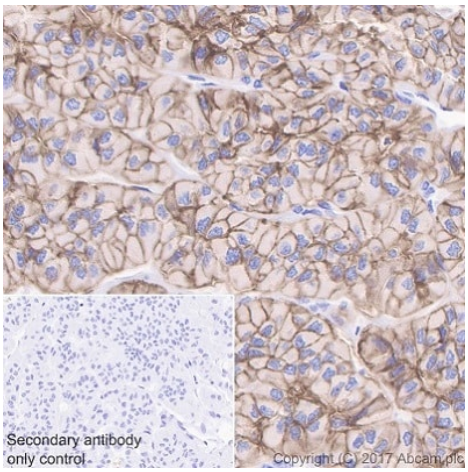
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 35 kDa
Observed band size: 40 kDa

Lanes 1 - 3: Merged signal (red and green). Green - ab223582 observed at 40 kDa. Red - loading control, **ab7291** (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

ab223582 was shown to react with EpCAM in A431 wild-type cells in Western blot. Loss of signal was observed when EpCAM knockout sample was used. A431 wild-type and EpCAM knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab223582 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 developed 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 hour at room temperature before imaging.

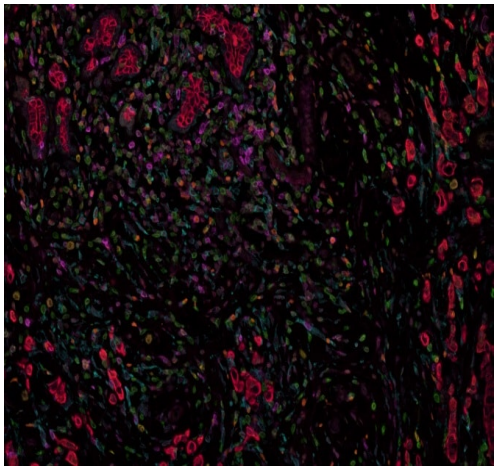


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [EPR20532-225] (ab223582)

Immunohistochemical analysis of paraffin-embedded human thyroid cancer tissue labeling EpCAM with ab223582 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Membranous staining on human thyroid cancer is observed (PMID: 15637741). Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Multiplex immunohistochemistry - Anti-EpCAM antibody [EPR20532-225] (ab223582)
This image is courtesy of ImmunoAtlas.

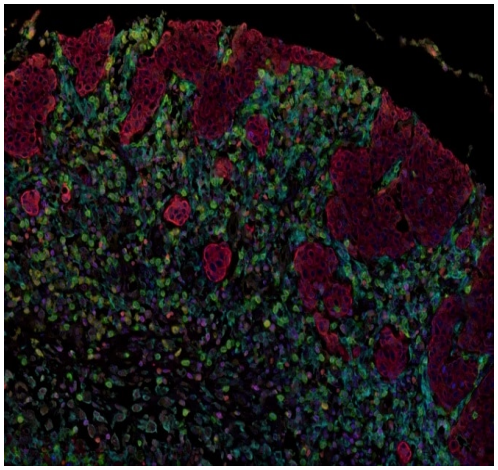
Fluorescence multiplex immunohistochemical analysis of Human breast cancer tissue (formalin-fixed paraffin-embedded section).

Merged staining of Anti-PD-L1 (**ab251611**; cyan; Opal™ 520), Anti-Granzyme B (**ab219803**; yellow; Opal™ 540), Anti-PD1 (**ab251613**; magenta; Opal™ 570), Anti-pan Cytokeratin (**ab264485**; red; Opal™ 620), Anti-EpCAM (**ab225894**; red; Opal™ 620), Anti-CD8 alpha (**ab251596**; green; Opal™ 650) and Anti-FOXP3 (**ab96048**; orange; Opal™ 690). EpCAM and pan-cytokeratin share the same dye and color. Dyes are pseudo-colored for better contrast of the markers.

The immunostaining was performed on a Leica Biosystems BOND® MAX instrument with an Opal™ 6-Plex Detection Kit (NEL821001KT, Akoya Biosciences®).

The section was incubated in six rounds of staining; sequentially for **ab251611** (1/750 dilution), **ab219803** (1/250 dilution), **ab251613** (1/750 dilution), **ab264485** (0.5 µg/ml), **ab225894** (1/1250 dilution), **ab251596** (1/1500 dilution) and **ab96048** (10 µg/ml); each using a separate fluorescent tyramide signal amplification system. EDTA based antigen retrieval (Leica Biosystems BOND® Epitope Retrieval Solution 2, pH 9.0, 20 minutes) was used in between rounds of tyramide signal amplification to remove the antibody from the previous round, to avoid any cross-reactivity. DAPI (dark blue) was used as a nuclear counter stain.
Microscopy and pseudocoloring of individual Opal™ dyes was performed using a Vectra 3 Imaging System (Akoya Biosciences®).

This data is courtesy of ImmunoAtlas and it can be found **here**.



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Fluorescence multiplex immunohistochemical analysis of Human breast cancer tissue (formalin-fixed paraffin-embedded section).

Merged staining of Anti-PD-L1 (**ab251611**), Anti-Granzyme B (**ab219803**), Anti-PD1 (**ab251613**), Anti-pan Cytokeratin (**ab264485**), Anti-EpCAM (**ab225894**), Anti-CD8 alpha (**ab251596**) and Anti-FOXP3 (**ab96048**).

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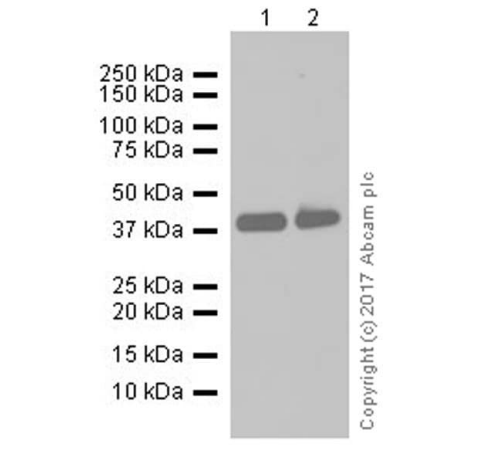


Western blot - Anti-EpCAM antibody [EPR20532-225] (ab223582)

All lanes : Anti-EpCAM antibody [EPR20532-225] (ab223582) at 1/5000 dilution

Lane 1 : HCT 116 (human colorectal carcinoma cell line) lysate
Lane 2 : HT-29 (human colorectal adenocarcinoma cell line) lysate

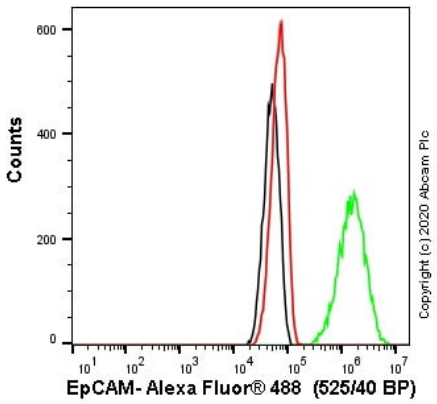
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 35 kDa
Observed band size: 42 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



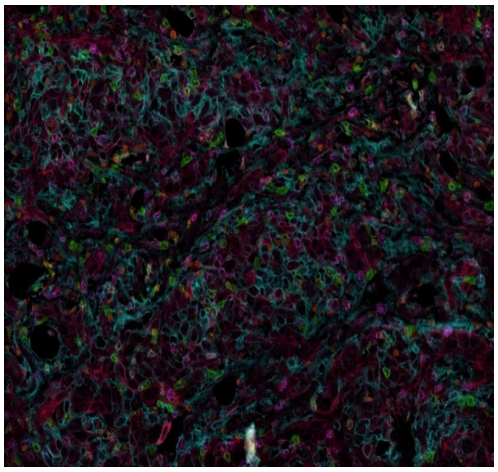
Flow Cytometry - Anti-EpCAM antibody [EPR20532-225] (ab223582)

Flow cytometry overlay histogram showing wild-type A431 (green line) and EPCAM knockout A431 cells stained with ab223582 (red line). The cells were incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab223582) (1x10^6 in 100µl at 0.2 µg/ml) for 30 min at 4°C.

The secondary antibody Goat anti-rabbit IgG H&L (Alexa Fluor® 488, pre-adsorbed) (**ab150081**) was used at 1/2000 for 30 min at 4°C.

Isotype control antibody was Rabbit IgG (monoclonal) (**ab172730**) used at the same concentration and conditions as the primary antibody (wild-type A431 - black line; EPCAM knockout A431 - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.



Multiplex immunohistochemistry - Anti-EpCAM antibody [EPR20532-225] (ab223582)

This image is courtesy of ImmunoAtlas.

Fluorescence multiplex immunohistochemical analysis of Human breast cancer tissue (formalin-fixed paraffin-embedded section).

Merged staining of Anti-PD-L1 (**ab251611**), Anti-Granzyme B (**ab219803**), Anti-PD1 (**ab251613**), Anti-pan Cytokeratin (**ab264485**), Anti-EpCAM (**ab225894**), Anti-CD8 alpha (**ab251596**) and Anti-FOXP3 (**ab96048**).

The immunostaining was performed on a Leica Biosystems BOND® MAX instrument with an Opal™ 6-Plex Detection Kit (NEL821001KT, Akoya Biosciences®).

The section was incubated in six rounds of staining; sequentially for **ab251611** (1/750 dilution), **ab219803** (1/250 dilution), **ab251613** (1/750 dilution), **ab264485** (0.5 µg/ml), **ab225894** (1/1250 dilution), **ab251596** (1/1500 dilution) and **ab96048** (10 µg/ml); each using a separate fluorescent tyramide signal amplification system. EDTA based antigen retrieval (Leica Biosystems BOND® Epitope Retrieval Solution 2, pH 9.0, 20 minutes) was used in between rounds of tyramide signal amplification to remove the antibody from the previous round, to avoid any cross-reactivity. DAPI (dark blue) was used as a nuclear counter stain.
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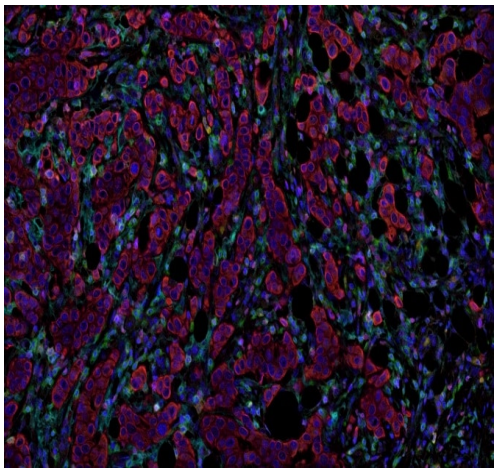

Multiplex immunohistochemistry - Anti-EpCAM antibody [EPR20532-225] (ab223582)

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Merged staining of Anti-PD-L1 (**ab251611**; cyan; Opal™ 520), Anti-Granzyme B (**ab219803**; yellow; Opal™ 540), Anti-PD1 (**ab251613**; magenta; Opal™ 570), Anti-pan Cytokeratin (**ab264485**; red; Opal™ 620), Anti-EpCAM (**ab225894**; red; Opal™ 620), Anti-CD8 alpha (**ab251596**; green; Opal™ 650) and Anti-FOXP3 (**ab96048**; orange; Opal™ 690). EpCAM and pan-cytokeratin share the same dye and color. Dyes are pseudo-colored for better contrast of the markers.

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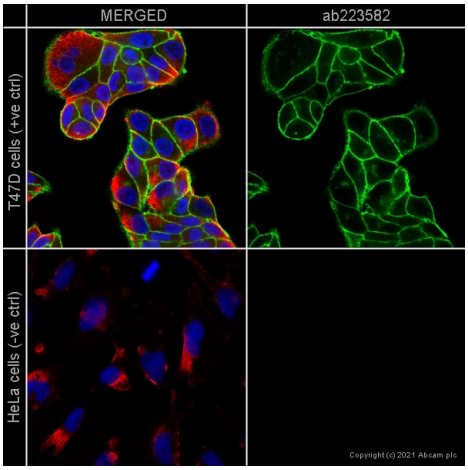
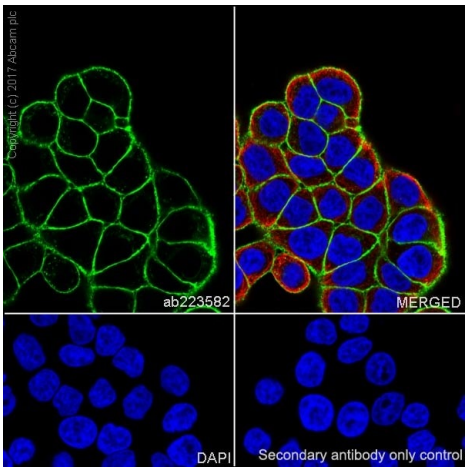


Immunocytochemistry/ Immunofluorescence - Anti-EpCAM antibody [EPR20532-225] (ab223582)

ab223582 staining EpCAM in T47D positive cells (top panel) and HeLa negative cells (bottom panel). The cells were fixed with 4% paraformaldehyde (10 min) then permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab223582 at 0.1µg/ml concentration and **ab7291** (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor® 594) (**ab150120**) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI. This antibody performed similarly using 100% methanol fixation. Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a single confocal section is

shown.

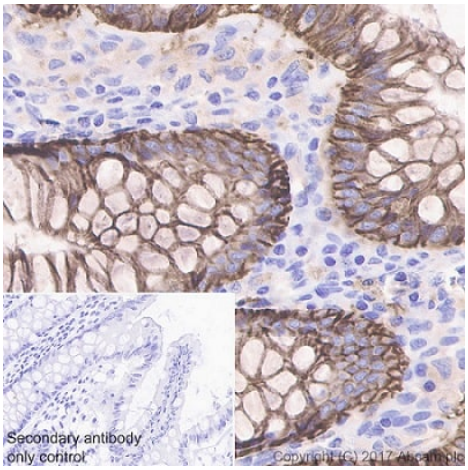


Immunocytochemistry/ Immunofluorescence - Anti-EpCAM antibody [EPR20532-225] (ab223582)

Immunofluorescent analysis of 4% paraformaledehyde-fixed, 0.1% Triton X-100 permeabilized HT-29 (human colorectal adenocarcinoma cell line) cells labeling EpCAM with ab223582 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing membranous staining on HT-29 cells.

The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (**ab195889**) (red) at 1/200 dilution.

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

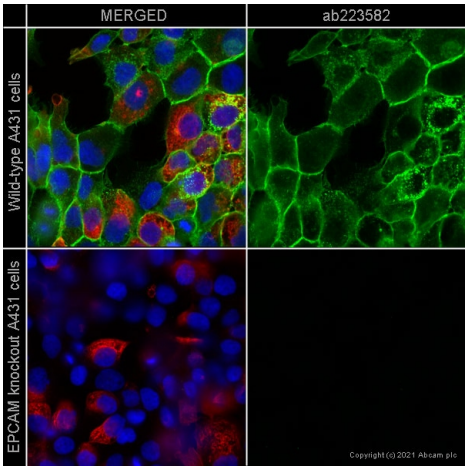


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-EpCAM antibody [EPR20532-225] (ab223582)

Immunohistochemical analysis of paraffin-embedded human colon tissue labeling EpCAM with ab223582 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Membranous staining on human colon is observed (PMID: 15637741). Counter stained with Hematoxylin.

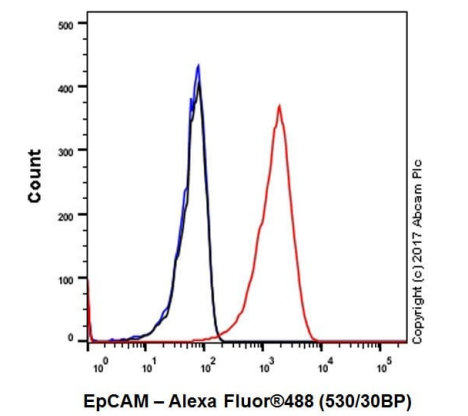
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-EpCAM antibody [EPR20532-225] (ab223582)

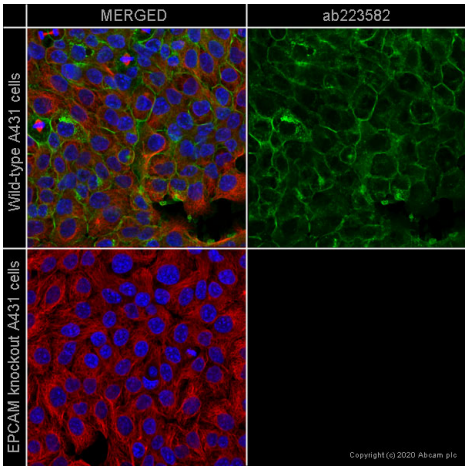
ab223582 staining EpCAM in wild-type A431 cells (top panel) and EpCAM knockout A431 cells (bottom panel). The cells were fixed with 4% paraformaldehyde (10 min) then permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab223582 at 0.1µg/ml concentration and **ab7291** (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor® 594) (**ab150120**) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI. This antibody performed similarly using 100% methanol fixation. Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a single confocal section is shown.



Flow Cytometry - Anti-EpCAM antibody [EPR20532-225] (ab223582)

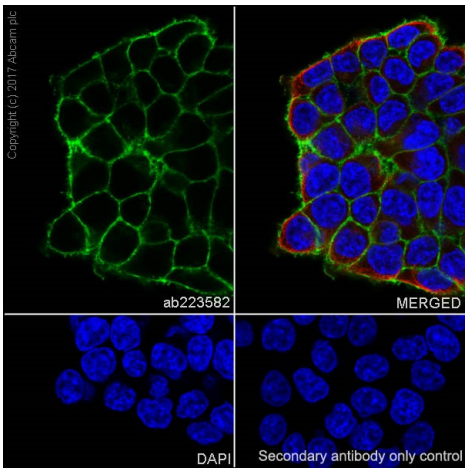
Flow cytometric analysis of HCT 116 (human colorectal carcinoma cell line) cell line labeling EpCAM with ab223582 at 1/500 (red) compared with a rabbit monoclonal IgG Isotype control (**ab172730**) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG (Alexa Fluor® 488) (**ab150077**), at 1/2000 dilution was used as the secondary antibody.

Total viable cells were gated for the final data.



Immunocytochemistry/ Immunofluorescence - Anti-EpCAM antibody [EPR20532-225] (ab223582)

ab223582 staining EpCAM in wild-type A431 cells (top panel) and EpCAM knockout A431 cells (bottom panel). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab223582 at 1/5000 dilution and **ab7291** (Mouse monoclonal to alpha Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse IgG (Alexa Fluor® 594) (**ab150120**) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI. Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).

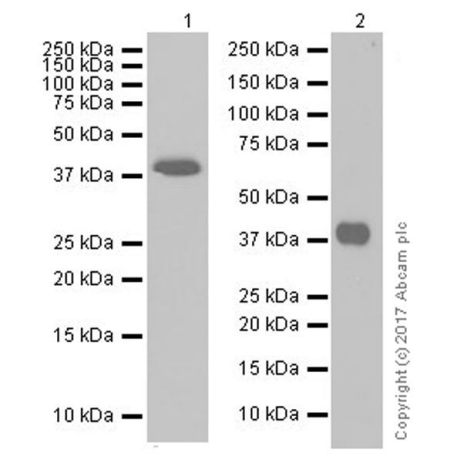


Immunocytochemistry/ Immunofluorescence - Anti-EpCAM antibody [EPR20532-225] (ab223582)

Immunofluorescent analysis of 4% paraformaledehyde-fixed, 0.1% Triton X-100 permeabilized HCT 116 (human colorectal carcinoma cell line) cells labeling EpCAM with ab223582 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing membranous staining on HT-29 cells.

The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (**ab195889**) (red) at 1/200 dilution.

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



Western blot - Anti-EpCAM antibody [EPR20532-225] (ab223582)

All lanes : Anti-EpCAM antibody [EPR20532-225] (ab223582) at 1/1000 dilution

Lane 1 : Human breat cancer lysate
Lane 2 : Human fetal kidney lysate

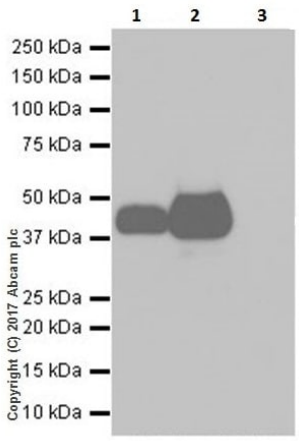
Lysates/proteins at 10 µg per lane.

Secondary
Lane 1 : VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at 1/1000 dilution
Lane 2 : VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at 1/4000 dilution

Predicted band size: 35 kDa
Observed band size: 42 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunoprecipitation - Anti-EpCAM antibody [EPR20532-225] (ab223582)

EpCAM was immunoprecipitated from 0.35 mg of HCT 116 (human colorectal carcinoma cell line) whole cell lysate with ab223582 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab223582 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

Lane 1: HCT 116 lysate 10 µg (Input).

Lane 2: ab223582 IP in HCT 116 lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab223582 in HCT 116 whole cell lysate.

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

Exposure time: 30 seconds.

Anti-EpCAM antibody [EPR20532-225] (ab223582)

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