# abcam

# Product datasheet

# Anti-EpCAM antibody [323/A3] ab85987



10 References 画像数 5

製品の概要

製品名 Anti-EpCAM antibody [323/A3]

製品の詳細 Mouse monoclonal [323/A3] to EpCAM

由来種 Mouse

特異性 We have data to indicate that this antibody may not cross react with Rat. However, this has not

been conclusively tested and expression levels may vary in certain cell lines/tissues.

アプリケーション 適用あり: ICC/IF, Flow Cyt, WB

種交差性 交差種: Human

免疫原 Tissue, cells or virus. MCF7 (human breast adenocarcinoma cell line) cells.

ポジティブ・コントロール Flow Cyt: A431 cells. ICC/IF: A431 and T47D cells. WB: MCF-7 cell lysate

特記事項

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

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid repeated freeze / thaw cycles.

**バッファー** pH: 7.40

Preservative: 0.097% Sodium azide

Constituent: PBS

精製度 Protein A purified

特記事項(精製) Purified from cell culture supernatant by protein A affinity chromatography, purity > 95% (by SDS-

PAGE).

ポリ/モノ モノクローナル

**クローン名** 323/A3

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#### アプリケーション

**The Abpromise guarantee** <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab85987の使用に適用されますアプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
ICC/IF		Use a concentration of 1 - 10 µg/ml.
Flow Cyt		Use a concentration of 1 - 4 μg/ml.
WB		Use a concentration of 1 - 2 µg/ml. Use under non reducing condition. Detects a band of approximately 40 kDa (predicted molecular weight: 35 kDa).

#### ターゲット情報

# 機能

#### 組織特異性

#### 関連疾患

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.

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.

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 extracolonic 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.

# 配列類似性

Belongs to the EPCAM family.

Contains 1 thyroglobulin type-1 domain.

翻訳後修飾

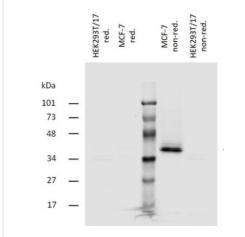
Hyperglycosylated in carcinoma tissue as compared with autologous normal epithelia.

Glycosylation at Asn-198 is crucial for protein stability.

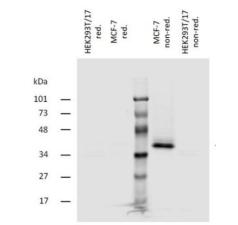
細胞内局在

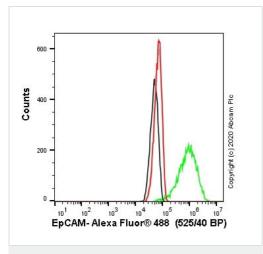
Lateral cell membrane. Cell junction > tight junction. Co-localizes with CLDN7 at the lateral cell membrane and tight junction.

#### 画像



Western blot - Anti-EpCAM antibody [323/A3] (ab85987)





Flow Cytometry - Anti-EpCAM antibody [323/A3] (ab85987)

All lanes: Anti-EpCAM antibody [323/A3] (ab85987) at 2 µg/ml

Lane 1: HEK293T/17 cell lysate (reducing)

Lane 2: MCF7 cell lysate (reducing)

Lane 3: HEK293T/17 cell lysate (non-reducing)

Lane 4: MCF7 cell lysate (non-reducing)

Secondary

All lanes: IRDye800-conjugated anti-mouse IgG1

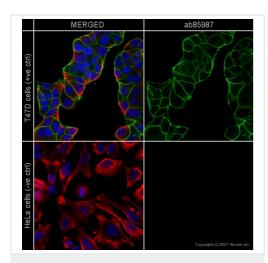
Predicted band size: 35 kDa Observed band size: 40 kDa

Flow cytometry overlay histogram showing wild-type A431 (green line) and EPCAM knockout A431 cells stained with ab85987 (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 (ab85987)  $(1x10^6 \text{ in } 100\mu\text{ at } 0.2 \,\mu\text{g/ml})$  for 30 min at 4°C.

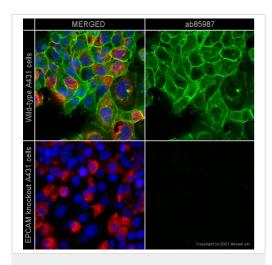
The secondary antibody Goat anti-mouse IgG H&L (Alexa Fluor® 488, pre-adsorbed) (ab150117) was used at 1/2000 for 30 min at 4°C.

Isotype control antibody was mouse IgG1κ (ab170190) used at the same concentration and conditions as the primary antibody (wildtype 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.



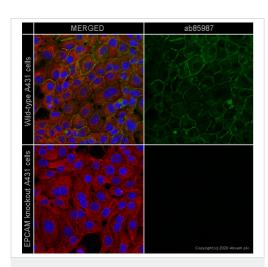
Immunocytochemistry/ Immunofluorescence - Anti-EpCAM antibody [323/A3] (ab85987)



Immunocytochemistry/ Immunofluorescence - Anti-EpCAM antibody [323/A3] (ab85987)

ab85987 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 ab85987 at 0.1µg/ml concentration and **ab6046** (Rabbit polyclonal to beta 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 mouse IgG (Alexa Fluor<sup>®</sup> 488) (**ab150117**) at 2 µg/ml (shown in green) and a goat secondary antibody to rabbit IgG (Alexa Fluor<sup>®</sup> 594) (**ab150080**) 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.

ab85987 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 ab85987 at 0.1µg/ml concentration and ab6046 (Rabbit polyclonal to beta 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 mouse IgG (Alexa Fluor<sup>®</sup> 488) (**ab150117**) at 2 μg/ml (shown in green) and a goat secondary antibody to rabbit lgG (Alexa Fluor® 594) (ab150080) 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 [323/A3] (ab85987)

ab85987 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 ab85987 at 0.5µg/ml concentration and ab6046 (Rabbit polyclonal to beta 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 mouse IgG (Alexa Fluor® 488) (ab150117) at 2 µg/ml (shown in green) and a goat secondary antibody to rabbit IgG (Alexa Fluor® 594) (ab150080) 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).

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