abcam

Product datasheet

Anti-E Cadherin antibody [M168] ab76055

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Overview

Product name	Anti-E Cadherin antibody [M168]
Description	Mouse monoclonal [M168] to E Cadherin
Specificity	Ab76055 does not crossreact with VE Cadherin or N Cadherin. This product may give a weak signal in Western Blot when using unstimulated cell lines. Abcam recommends using A431 cells treated with pervandate (1mM, 30 minutes) as a positive control. Abcam recommends ab40772 and ab11512 as a alternative when using unstimulated samples in Western Blot.
Tested applications	**Suitable for:** Flow Cyt, ICC/IF, IHC-P, IHC-Fr, WB, IP, ELISA, ICC
Species reactivity	**Reacts with:** Mouse, Rat, Horse, Human
Immunogen	Recombinant fragment containing amino acids in the C terminal region of Mouse E Cadherin. This sequence is highly conserved in human and rat E Cadherin.
Positive control	WB: Human A431 cells treated with pervanadate (1 mM) for 30 min. IHC-P: human normal colon tissue sections IF/ICC: A431 cell line

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer	Preservative: 0.05% Sodium Azide Constituents: 50% Glycerol, PBS, 1mg/ml BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	M168
Isotype	IgG1

Applications

Our Abpromise guarantee covers the use of **ab76055** 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

Application	Abreviews	Notes
Flow Cyt		1/100. ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★☆	Use at an assay dependent concentration. PubMed: 23405249
IHC-P	★★★★★	Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
IHC-Fr		Use at an assay dependent concentration. PubMed: 21769484
WB	★★★★☆	1/100 - 1/1000. Predicted molecular weight: 97 kDa. This product may give a weak signal in Western Blot when using unstimulated cell lines. Abcam recommends using A431 cells treated with pervandate (1mM, 30 minutes) as a positive control.
IP		1/100.
ELISA		1/2000.
ICC		1/250.

Target

Function

Cadherins are calcium-dependent cell adhesion proteins. They preferentially interact with themselves in a homophilic manner in connecting cells; cadherins may thus contribute to the sorting of heterogeneous cell types. CDH1 is involved in mechanisms regulating cell-cell adhesions, mobility and proliferation of epithelial cells. Has a potent invasive suppressor role. It is a ligand for integrin alpha-E/beta-7.
E-Cad/CTF2 promotes non-amyloidogenic degradation of Abeta precursors. Has a strong inhibitory effect on APP C99 and C83 production.

Tissue specificity

Non-neural epithelial tissues.

Involvement in disease

Defects in CDH1 are the cause of hereditary diffuse gastric cancer (HDGC) [MIM:137215]. An autosomal dominant cancer predisposition syndrome with increased susceptibility to diffuse gastric cancer. Diffuse gastric cancer is a malignant disease characterized by poorly differentiated infiltrating lesions resulting in thickening of the stomach. Malignant tumors start in the stomach, can spread to the esophagus or the small intestine, and can extend through the stomach wall to nearby lymph nodes and organs. It also can metastasize to other parts of the body. Note=Heterozygous germline mutations CDH1 are responsible for familial cases of diffuse gastric cancer. Somatic mutations in the has also been found in patients with sporadic diffuse gastric cancer and lobular breast cancer.
Defects in CDH1 are a cause of susceptibility to endometrial cancer (ENDMC) [MIM:608089].
Defects in CDH1 are a cause of susceptibility to ovarian cancer (OC) [MIM:167000]. Ovarian cancer common malignancy originating from ovarian tissue. Although many histologic types of ovarian neoplasms have been described, epithelial ovarian carcinoma is the most common form. Ovarian cancers are often asymptomatic and the recognized signs and symptoms, even of late-stage disease, are vague. Consequently, most patients are diagnosed with advanced disease.

Sequence similarities

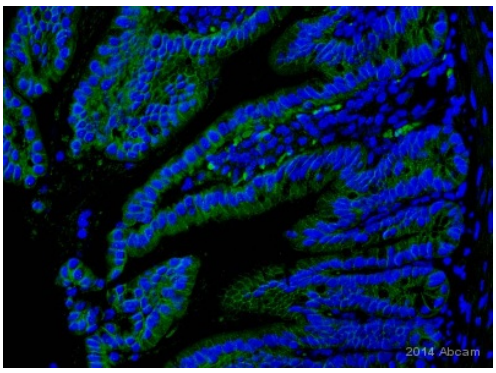
Contains 5 cadherin domains.

Post-translational modifications

During apoptosis or with calcium influx, cleaved by a membrane-bound metalloproteinase (ADAM10), PS1/gamma-secretase and caspase-3 to produce fragments of about 38 kDa (E-CAD/CTF1), 33 kDa (E-CAD/CTF2) and 29 kDa (E-CAD/CTF3), respectively. Processing by

	the metalloproteinase, induced by calcium influx, causes disruption of cell-cell adhesion and the subsequent release of beta-catenin into the cytoplasm. The residual membrane-tethered cleavage product is rapidly degraded via an intracellular proteolytic pathway. Cleavage by caspase-3 releases the cytoplasmic tail resulting in disintegration of the actin microfilament system. The gamma-secretase-mediated cleavage promotes disassembly of adherens junctions.
Cellular localization	Cell junction. Cell membrane. Endosome. Golgi apparatus > trans-Golgi network. Colocalizes with DLGAP5 at sites of cell-cell contact in intestinal epithelial cells. Anchored to actin microfilaments through association with alpha-, beta- and gamma-catenin. Sequential proteolysis induced by apoptosis or calcium influx, results in translocation from sites of cell-cell contact to the cytoplasm. Colocalizes with RAB11A endosomes during its transport from the Golgi apparatus to the plasma membrane.

Anti-E Cadherin antibody [M168] images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-E Cadherin [M168] antibody (ab76055)

This image is courtesy of an anonymous Abreview

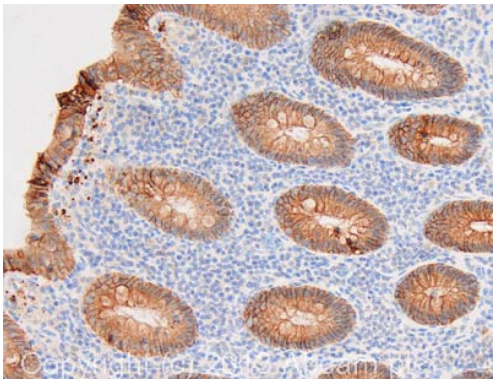
ab76055 staining E Cadherin in mouse intestine tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with paraformaldehyde and blocked with 0.1% goat Fab anti-mouse IgG for 30 minutes at 25°C; antigen retrieval was by heat mediation in 10mM citrate buffer, pH 6. Samples were incubated with primary antibody (1/250) for 2 hours at 25°C. An Alexa Fluor® 488-conjugated donkey anti-mouse IgG polyclonal (1/500) was used as the secondary antibody.

Western blot - Anti-E Cadherin antibody [M168] (ab76055)

All lanes : Anti-E Cadherin antibody [M168] (ab76055) at 1/1000 dilution

Lane 1 : Human A431 cells treated with pervanadate (1 mM) for 30 min
Lane 2 : Human A431 cells treated with pervanadate (1 mM) for 30 min, then treated with akaline phosphatase

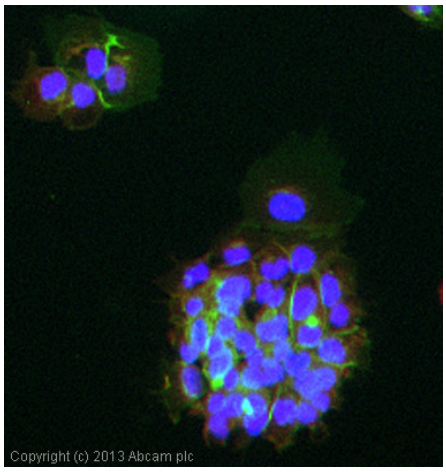
Predicted band size : 97 kDa
Observed band size : 120 kDa
Membranes were incubated with diluted antibody in 5% non-fat milk, PBS, 0.04% Tween20 for 1hour at room temperature.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-E Cadherin antibody [M168] (ab76055)

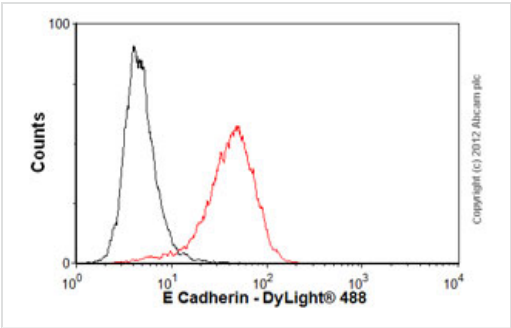
IHC image of E Cadherin staining in human normal colon formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab76055, 5µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunocytochemistry/ Immunofluorescence - Anti-E Cadherin [M168] antibody (ab76055)

ICC/IF image of ab76055 stained A431 cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab76055, 1/100 dilution) overnight at +4°C. The secondary antibody (green) was ab96879, DyLight® 488 goat anti-mouse IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Flow Cytometry - Anti-E Cadherin antibody [M168] (ab76055)

Overlay histogram showing A431 cells stained with ab76055 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab76055, 1/100 dilution) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22ºC. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in A431 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.