

# Anti-MUC1 antibody [SM3] ab22711

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### Overview

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<b>Product name</b>	Anti-MUC1 antibody [SM3]
<b>Description</b>	Mouse monoclonal [SM3] to MUC1
<b>Host species</b>	Mouse
<b>Tested applications</b>	<b>Suitable for:</b> Flow Cyt (Intra), ICC/IF, IHC-P <b>Unsuitable for:</b> WB
<b>Species reactivity</b>	<b>Reacts with:</b> Human
<b>Immunogen</b>	Full length protein. This information is proprietary to Abcam and/or its suppliers.
<b>Positive control</b>	Flow Cyt: MCF7 cells. IHC-P: Human breast carcinoma tissue. ICC/IF: MCF7 cell line.
<b>General notes</b>	<p>This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact <a href="mailto:orders@abcam.com">orders@abcam.com</a>.</p> <p>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.</p> <p>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&amp;As</p>

### Properties

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<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
<b>Storage buffer</b>	pH: 7.40 Preservative: 0.02% Sodium azide Constituent: PBS
<b>Purity</b>	Protein G purified
<b>Clonality</b>	Monoclonal
<b>Clone number</b>	SM3
<b>Isotype</b>	IgG1

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab22711 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 (Intra)		Use 1 µg for 10 <sup>6</sup> cells.
ICC/IF		Use a concentration of 1 µg/ml.
IHC-P	★★★★★ (1)	Use a concentration of 5 - 10 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

**Application notes** Is unsuitable for WB.

## Target

<b>Function</b>	<p>The alpha subunit has cell adhesive properties. Can act both as an adhesion and an anti-adhesion protein. May provide a protective layer on epithelial cells against bacterial and enzyme attack.</p> <p>The beta subunit contains a C-terminal domain which is involved in cell signaling, through phosphorylations and protein-protein interactions. Modulates signaling in ERK, SRC and NF-kappa-B pathways. In activated T-cells, influences directly or indirectly the Ras/MAPK pathway. Promotes tumor progression. Regulates TP53-mediated transcription and determines cell fate in the genotoxic stress response. Binds, together with KLF4, the PE21 promoter element of TP53 and represses TP53 activity.</p>
<b>Tissue specificity</b>	<p>Expressed on the apical surface of epithelial cells, especially of airway passages, breast and uterus. Also expressed in activated and unactivated T-cells. Overexpressed in epithelial tumors, such as breast or ovarian cancer and also in non-epithelial tumor cells. Isoform Y is expressed in tumor cells only.</p>
<b>Involvement in disease</b>	<p>MUC1/CA 15-3 is used as a serological clinical marker of breast cancer to monitor response to breast cancer treatment and disease recurrence (PubMed:20816948). Decreased levels over time may be indicative of a positive response to treatment. Conversely, increased levels may indicate disease progression. At an early stage disease, only 21% of patients exhibit high MUC1/CA 15-3 levels, that is why CA 15-3 is not a useful screening test. Most antibodies target the highly immunodominant core peptide domain of 20 amino acid (APDTRPAPGSTAPPAHGVTS) tandem repeats. Some antibodies recognize glycosylated epitopes.</p> <p>Medullary cystic kidney disease 1</p>
<b>Sequence similarities</b>	<p>Contains 1 SEA domain.</p>
<b>Developmental stage</b>	<p>During fetal development, expressed at low levels in the colonic epithelium from 13 weeks of gestation.</p>
<b>Post-translational modifications</b>	<p>Highly glycosylated (N- and O-linked carbohydrates and sialic acid). O-glycosylated to a varying degree on serine and threonine residues within each tandem repeat, ranging from mono- to penta-glycosylation. The average density ranges from about 50% in human milk to over 90% in T47D breast cancer cells. Further sialylation occurs during recycling. Membrane-shed</p>

glycoproteins from kidney and breast cancer cells have preferentially sialylated core 1 structures, while secreted forms from the same tissues display mainly core 2 structures. The O-glycosylated content is overlapping in both these tissues with terminal fucose and galactose, 2- and 3-linked galactose, 3- and 3,6-linked GalNAc-ol and 4-linked GlcNAc predominating. Differentially O-glycosylated in breast carcinomas with 3,4-linked GlcNAc. N-glycosylation consists of high-mannose, acidic complex-type and hybrid glycans in the secreted form MUC1/SEC, and neutral complex-type in the transmembrane form, MUC1/TM.

Proteolytic cleavage in the SEA domain occurs in the endoplasmic reticulum by an autoproteolytic mechanism and requires the full-length SEA domain as well as requiring a Ser, Thr or Cys residue at the P + 1 site. Cleavage at this site also occurs on isoform MUC1/X but not on isoform MUC1/Y. Ectodomain shedding is mediated by ADAM17.

Dual palmitoylation on cysteine residues in the CQC motif is required for recycling from endosomes back to the plasma membrane.

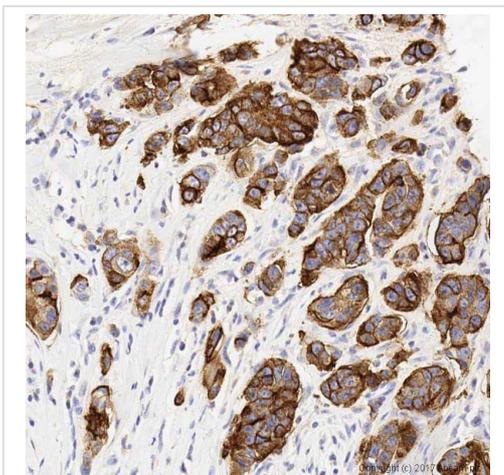
Phosphorylated on tyrosines and serine residues in the C-terminal. Phosphorylation on tyrosines in the C-terminal increases the nuclear location of MUC1 and beta-catenin. Phosphorylation by PKC delta induces binding of MUC1 to beta-catenin/CTNNB1 and thus decreases the formation of the beta-catenin/E-cadherin complex. Src-mediated phosphorylation inhibits interaction with GSK3B. Src- and EGFR-mediated phosphorylation on Tyr-1229 increases binding to beta-catenin/CTNNB1. GSK3B-mediated phosphorylation on Ser-1227 decreases this interaction but restores the formation of the beta-cadherin/E-cadherin complex. On T-cell receptor activation, phosphorylated by LCK. PDGFR-mediated phosphorylation increases nuclear colocalization of MUC1CT and CTNNB1.

The N-terminal sequence has been shown to begin at position 24 or 28.

## Cellular localization

Secreted; Cell membrane. Cytoplasm. Nucleus. On EGF and PDGFRB stimulation, transported to the nucleus through interaction with CTNNB1, a process which is stimulated by phosphorylation. On HRG stimulation, colocalizes with JUP/gamma-catenin at the nucleus and Apical cell membrane. Exclusively located in the apical domain of the plasma membrane of highly polarized epithelial cells. After endocytosis, internalized and recycled to the cell membrane. Located to microvilli and to the tips of long filopodial protrusions.

## Images

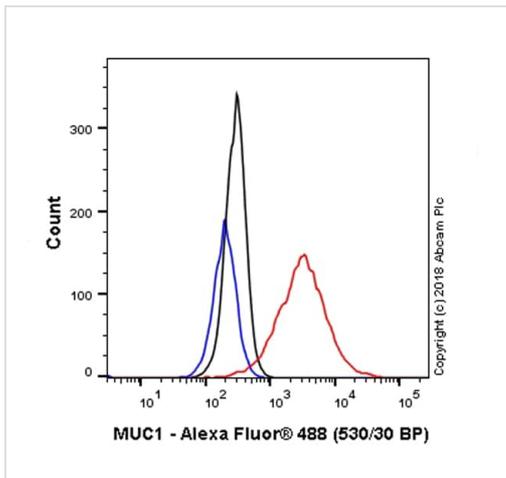


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MUC1 antibody [SM3] (ab22711)

IHC image of MUC1 staining in a formalin fixed, paraffin embedded human breast carcinoma 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 ab22711, 10 µg/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.

\*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



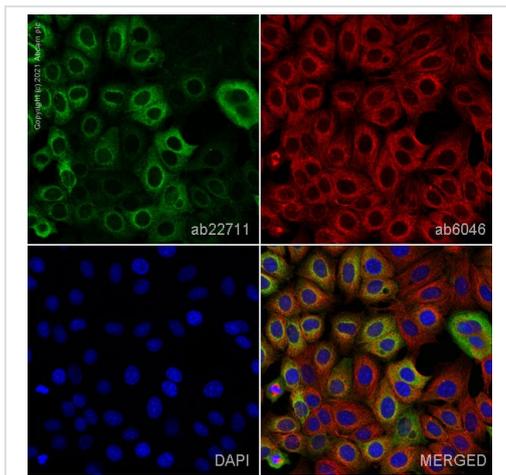
Flow Cytometry (Intracellular) - Anti-MUC1 antibody [SM3] (ab22711)

Overlay histogram showing MCF7 cells stained with ab22711 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab22711, 1µg/1x10<sup>6</sup> cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-mouse IgG (H&L) (**ab150117**) at 1/2000 dilution for 30 min at 22°C.

Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (**ab170190**, 1µg/1x10<sup>6</sup> cells) used under the same conditions.

Acquisition of >5,000 events was performed.

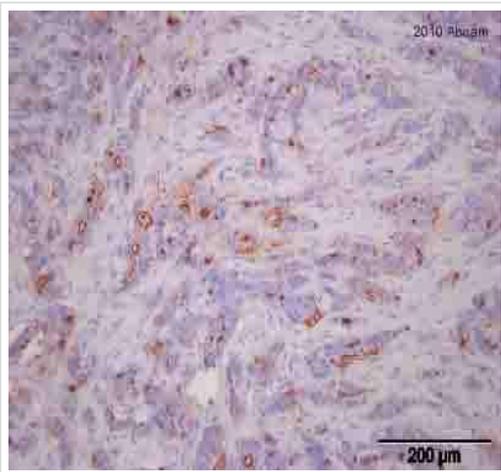
This antibody gave a positive signal in MCF7 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Triton X-100 for 15 min used under the same conditions.



Immunocytochemistry/ Immunofluorescence - Anti-MUC1 antibody [SM3] (ab22711)

ab22711 staining MUC1 in MCF7 cells. The cells were fixed with 100% methanol (5 min), 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 overnight at 4°C with ab22711 at 1µg/ml and **ab6046**, Rabbit polyclonal to beta Tubulin - Loading Control. Cells were then incubated with **ab150117**, Goat polyclonal Secondary Antibody to Mouse IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and **ab150080**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 594) at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MUC1 antibody [SM3] (ab22711)

Image courtesy of an anonymous Abreview.

ab22711 staining MUC1 in human breast carcinoma tissue sections by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Tissue was fixed with formaldehyde and a heat mediated antigen retrieval step was performed using 10mM citrate buffer. The primary antibody was used undiluted for 18 hours at 4°C. An undiluted HRP-conjugated goat anti-mouse IgG polyclonal was used as secondary antibody.

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