abcam

Product datasheet

Anti-BRCA1 antibody [MS13] ab16781

★★★★★ <u>5 Abreviews</u> <u>13 References</u> 4 Images

Overview

Product name Anti-BRCA1 antibody [MS13]

Description Mouse monoclonal [MS13] to BRCA1

Host species Mouse

Tested applications Suitable for: Flow Cyt (Intra), ICC/IF, IHC-P

Species reactivity Reacts with: Human

Predicted to work with: African green monkey

Immunogen Recombinant full length protein corresponding to Human BRCA1.

Epitope Within the N-terminal 304 amino acids of BRCA1.

Positive control IHC-P: Normal breast tissue. ICC/IF: MCF7 cells. Flow Cyt (Intra): MCF7 cells.

General notes

This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

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

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 Preservative: 0.02% Sodium azide

Constituents: PBS, 6.97% L-Arginine

Purity Protein G purified

Clonality Monoclonal

Clone number MS13

1

MyelomaNS1IsotypeIgG1Light chain typekappa

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab16781 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 1µg for 10 ⁶ cells. ab170190 - Mouse monoclonal lgG1, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★ <u>(3)</u>	Use a concentration of 1 µg/ml.
IHC-P	★★★★ (1)	Use a concentration of 5 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Target

Function

E3 ubiquitin-protein ligase that specifically mediates the formation of 'Lys-6'-linked polyubiquitin chains and plays a central role in DNA repair by facilitating cellular responses to DNA damage. It is unclear whether it also mediates the formation of other types of polyubiquitin chains. The E3 ubiquitin-protein ligase activity is required for its tumor suppressor function. The BRCA1-BARD1 heterodimer coordinates a diverse range of cellular pathways such as DNA damage repair, ubiquitination and transcriptional regulation to maintain genomic stability. Regulates centrosomal microtubule nucleation. Required for normal cell cycle progression from G2 to mitosis. Required for appropriate cell cycle arrests after ionizing irradiation in both the S-phase and the G2 phase of the cell cycle. Involved in transcriptional regulation of P21 in response to DNA damage. Required for FANCD2 targeting to sites of DNA damage. May function as a transcriptional regulator. Inhibits lipid synthesis by binding to inactive phosphorylated ACACA and preventing its dephosphorylation. Contributes to homologous recombination repair (HRR) via its direct interaction with PALB2, fine-tunes recombinational repair partly through its modulatory role in the PALB2-dependent loading of BRCA2-RAD51 repair machinery at DNA breaks.

Tissue specificity

lsoform 1 and isoform 3 are widely expressed. Isoform 3 is reduced or absent in several breast and ovarian cancer cell lines.

Pathway

Protein modification; protein ubiquitination.

Involvement in disease

Defects in BRCA1 are a cause of susceptibility to breast cancer (BC) [MIM:114480]. A common malignancy originating from breast epithelial tissue. Breast neoplasms can be distinguished by their histologic pattern. Invasive ductal carcinoma is by far the most common type. Breast cancer is etiologically and genetically heterogeneous. Important genetic factors have been indicated by familial occurrence and bilateral involvement. Mutations at more than one locus can be involved in different families or even in the same case. Note=Mutations in BRCA1 are thought to be responsible for 45% of inherited breast cancer. Moreover, BRCA1 carriers have a 4-fold increased risk of colon cancer, whereas male carriers face a 3-fold increased risk of prostate

cancer. Cells lacking BRCA1 show defects in DNA repair by homologous recombination. Defects in BRCA1 are a cause of susceptibility to breast-ovarian cancer familial type 1 (BROVCA1) [MIM:604370]. A condition associated with familial predisposition to cancer of the breast and ovaries. Characteristic features in affected families are an early age of onset of breast cancer (often before age 50), increased chance of bilateral cancers (cancer that develop in both breasts, or both ovaries, independently), frequent occurrence of breast cancer among men, increased incidence of tumors of other specific organs, such as the prostate. Note=Mutations in BRCA1 are thought to be responsible for more than 80% of inherited breast-ovarian cancer. Defects in BRCA1 are a cause of genetic susceptibility to ovarian cancer [MIM:113705].

Sequence similarities

Contains 2 BRCT domains.

Contains 1 RING-type zinc finger.

Domain

The BRCT domains recognize and bind phosphorylated pSXXF motif on proteins. The interaction with the phosphorylated pSXXF motif of FAM175A/Abraxas, recruits BRCA1 at DNA damage sites.

The RING-type zinc finger domain interacts with BAP1.

Post-translational modifications

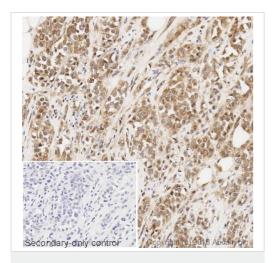
Phosphorylation at Ser-308 by STK6/AURKA is required for normal cell cycle progression from G2 to mitosis. Phosphorylated in response to IR, UV, and various stimuli that cause checkpoint activation, probably by ATM or ATR.

Autoubiquitinated, undergoes 'Lys-6'-linked polyubiquitination. 'Lys-6'-linked polyubiquitination does not promote degradation.

Cellular localization

Cytoplasm; Nucleus. Localizes at sites of DNA damage at double-strand breaks (DSBs) and recruitment to DNA damage sites is mediated by the BRCA1-A complex.

Images

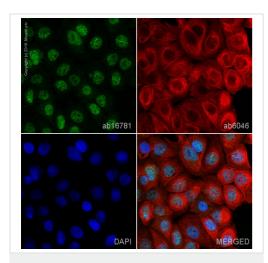


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-BRCA1 antibody [MS13] (ab16781)

IHC image of BRCA1 staining in a section of formalin-fixed paraffinembedded normal human breast* performed on a Leica BONDTM 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 20mins. The section was then incubated with ab16781, 5ug/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. The inset secondary-only control image is taken from an identical assay without primary antibody.

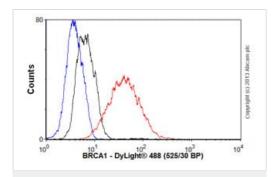
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

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



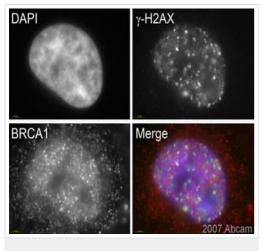
Immunocytochemistry/ Immunofluorescence - Anti-BRCA1 antibody [MS13] (ab16781)

ab16781 staining BRCA1 in MCF7 cells. The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% Triton X-100 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 ab16781 at 1µg/ml concentration and ab6046 at 1/1000 dilution overnight at +4°C, followed by a further incubation at room temperature for 1h with Goat Anti-Mouse lgG H&L (ab150117) at 1/1000 dilution (shown in green) and Goat anti-Rabbit lgG (ab150080) at 1/1000 dilution (shown in pseudocolor red). Nuclear DNA was labelled in blue with DAPI.



Flow Cytometry (Intracellular) - Anti-BRCA1 antibody [MS13] (ab16781)

Overlay histogram showing MCF7 cells stained with ab16781 (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 (ab16781, 1µg/1x10⁶ cells) for 30 min at 22°C. The secondary antibody used was DyLight[®] 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line). Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in MCF7 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



ab16781 (1/200) detecting BRCA1 in HeLa cells in conjunction with a goat anti-mouse secondary antibody conjugated to Cy3 (red). Cells were also counterstained with DAPI in order to highlight the nucleus (blue), treated with Bleomycin and incubated with an antibody against Histone H2AX in order to create and expose DNA double strand breaks (green). Please refer to abreviews for further details.

Immunocytochemistry/ Immunofluorescence - Anti-BRCA1 antibody [MS13] (ab16781)

This image is courtesy of an Abreview submitted by Dr Kirk McManus

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