abcam

Product datasheet

Anti-ATM antibody [Y170] ab32420



★★★★★ 8 Abreviews 118 References 画像数 12

製品の概要

製品名 Anti-ATM antibody [Y170]

製品の詳細 Rabbit monoclonal [Y170] to ATM

由来種 Rabbit

アプリケーション 適用あり: Flow Cyt (Intra), WB, IHC-P, ICC/IF

適用なし: IP

種交差性 交差種: Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: 293 cell lysate. Flow Cyt (intra): HeLa cells ICC/IF: HeLa and HepG2 cells

特記事項 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.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

精製度 Protein A purified

ポリモノ モノクローナル

クローン名 Y170 アイソタイプ lqG

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ab32420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
Flow Cyt (Intra)		1/20.
WB	★★★★★ (5)	1/1000 - 1/10000. Predicted molecular weight: 350 kDa.
IHC-P	★★★★★ (3)	1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/250 - 1/500.

追加情報

Is unsuitable for IP.

ターゲット情報

機能

Serine/threonine protein kinase which activates checkpoint signaling upon double strand breaks (DSBs), apoptosis and genotoxic stresses such as ionizing ultraviolet A light (UVA), thereby acting as a DNA damage sensor. Recognizes the substrate consensus sequence [ST]-Q. Phosphorylates 'Ser-139' of histone variant H2AX/H2AFX at double strand breaks (DSBs), thereby regulating DNA damage response mechanism. Also plays a role in pre-B cell allelic exclusion, a process leading to expression of a single immunoglobulin heavy chain allele to enforce clonality and monospecific recognition by the B-cell antigen receptor (BCR) expressed on individual B lymphocytes. After the introduction of DNA breaks by the RAG complex on one immunoglobulin allele, acts by mediating a repositioning of the second allele to pericentromeric heterochromatin, preventing accessibility to the RAG complex and recombination of the second allele. Also involved in signal transduction and cell cycle control. May function as a tumor suppressor. Necessary for activation of ABL1 and SAPK. Phosphorylates p53/TP53, FANCD2, NFKBIA, BRCA1, CTIP, nibrin (NBN), TERF1, RAD9 and DCLRE1C. May play a role in vesicle and/or protein transport. Could play a role in T-cell development, gonad and neurological function. Plays a role in replication-dependent histone mRNA degradation. Binds DNA ends.

組織特異性

関連疾患

Found in pancreas, kidney, skeletal muscle, liver, lung, placenta, brain, heart, spleen, thymus, testis, ovary, small intestine, colon and leukocytes.

Defects in ATM are the cause of ataxia telangiectasia (AT) [MIM:208900]; also known as Louis-Bar syndrome, which includes four complementation groups: A, C, D and E. This rare recessive disorder is characterized by progressive cerebellar ataxia, dilation of the blood vessels in the conjunctiva and eyeballs, immunodeficiency, growth retardation and sexual immaturity. AT patients have a strong predisposition to cancer; about 30% of patients develop tumors, particularly lymphomas and leukemias. Cells from affected individuals are highly sensitive to damage by ionizing radiation and resistant to inhibition of DNA synthesis following irradiation. Note=Defects in ATM contribute to T-cell acute lymphoblastic leukemia (TALL) and T-prolymphocytic leukemia (TPLL). TPLL is characterized by a high white blood cell count, with a predominance of prolymphocytes, marked splenomegaly, lymphadenopathy, skin lesions and serous effusion. The clinical course is highly aggressive, with poor response to chemotherapy and short survival time. TPLL occurs both in adults as a sporadic disease and in younger AT patients.

Note=Defects in ATM contribute to B-cell non-Hodgkin lymphomas (BNHL), including mantle cell

lymphoma (MCL).

Note=Defects in ATM contribute to B-cell chronic lymphocytic leukemia (BCLL). BCLL is the commonest form of leukemia in the elderly. It is characterized by the accumulation of mature CD5+ B lymphocytes, lymphadenopathy, immunodeficiency and bone marrow failure.

配列類似性 Belongs to the PI3/PI4-kinase family. ATM subfamily.

Contains 1 FAT domain.
Contains 1 FATC domain.
Contains 1 PI3K/PI4K domain.

The FATC domain is required for interaction with KAT5.

翻訳後修飾 Phosphorylated by NUAK1/ARK5. Autophosphorylation on Ser-367, Ser-1893, Ser-1981

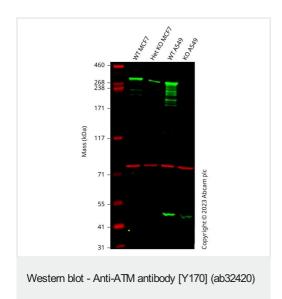
correlates with DNA damage-mediated activation of the kinase.

Acetylation, on DNA damage, is required for activation of the kinase activity, dimer-monomer transition, and subsequent autophosphorylation on Ser-1981. Acetylated in vitro by KAT5/TIP60.

細胞内局在 Nucleus. Cytoplasmic vesicle. Primarily nuclear. Found also in endocytic vesicles in association

with beta-adaptin.

画像



All lanes: Anti-ATM antibody [Y170] (ab32420) at 1/1000 dilution

Lane 1: Wild-type MCF7 cell lysate

Lane 2: ATM knockout MCF7 cell lysate

Lane 3: Wild-type A549 cell lysate

Lane 4: ATM knockout A549 ab283811 cell lysate

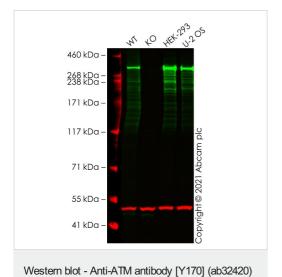
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 350 kDa Observed band size: 350 kDa

Western blot: Anti-ATM antibody [Y170] (ab32420) staining at 1/1000 dilution, shown in green; Mouse anti-CANX [CANX/1543] (ab238078) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32420 was shown to bind specifically to ATM. A band was observed at 350 kDa in wild-type MCF7 cell lysates with a reduction in signal observed at this size in ATM heterozygous knockout cell line ab282630. To generate this image, wild-type and ATM heterozygous knockout MCF7 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were

blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit lgG H&L 800CW and Goat anti-Mouse lgG H&L 680RD at 1/20000 dilution.



All lanes: Anti-ATM antibody [Y170] (ab32420) at 1/1000 dilution

Lane 1: Wild-type A549 cell lysate

Lane 2: ATM knockout A549 cell lysate

Lane 3: HEK-293 cell lysate

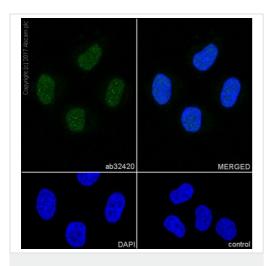
Lane 4: U-2 OS cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

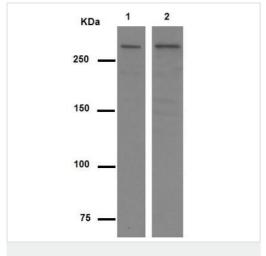
Predicted band size: 350 kDa **Observed band size:** 350 kDa

False colour image of Western blot: Anti-ATM antibody [Y170] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32420 was shown to bind specifically to ATM. A band was observed at 350 kDa in wild-type A549 cell lysates with no signal observed at this size in ATM knockout cell line ab276095 (knockout cell lysate ab283834). To generate this image, wild-type and ATM knockout A549 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 5% milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDve® 680RD) preabsorbed (ab216776) at 1/20000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-ATM antibody [Y170] (ab32420)

Immunocytochemistry/ Immunofluorescence analysis of HeLa cells labeling ATM with ab32420 at 1/500. Goat anti rabbit IgG(Alexa Fluor[®] 488), <u>ab150077</u> at 1/1000 was used as the secondary antibody. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. Nulei were counterstained blue with DAPI.



Western blot - Anti-ATM antibody [Y170] (ab32420)

All lanes : Anti-ATM antibody [Y170] (ab32420) at 1/3000 dilution (Purified)

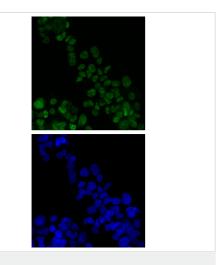
Lane 1 : HeLa cell lysate
Lane 2 : HEK293 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

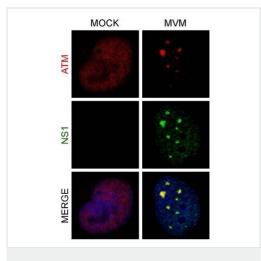
All lanes: HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 350 kDa **Observed band size:** 370 kDa



Immunocytochemistry/ Immunofluorescence - Anti-ATM antibody [Y170] (ab32420)

Immunocytochemistry/ Immunofluorescence analysis of HepG2 cells labeling ATM with ab32420 at 1/500. Goat anti rabbit lgG(Alexa Fluor[®] 488), **ab150077** at 1/1000 was used as the secondary antibody. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. Nulei were counterstained blue with DAPI.



Immunocytochemistry/ Immunofluorescence - Anti-ATM antibody [Y170] (ab32420)

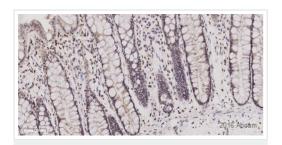
Adeyemi, R.O. et al PLoS Pathog. 2010 Oct 7;6(10):e1001141. doi: 10.1371/journal.ppat.1001141 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

DNA repair proteins accumulate at MVM APAR bodies

Repair proteins accumulate at APAR bodies. NB324K cells were infected with MVMp (MOI of 10) for 16 hr before being fixed and processed for immunofluorescence. Cells were stained with the indicated antibodies to mark DDR repair proteins. APAR bodies were detected with antibodies to NS1. Nuclei were stained with DAPI. All images were captured using an objective of 63×.

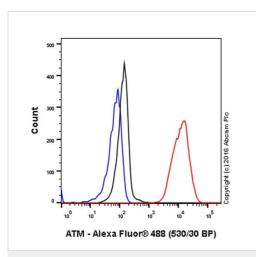
Cells were fixed with 4% paraformaldehyde for 15 minutes and permeabilized with 0.5% Triton X-100 in PBS for 15 minutes.

(Image shows the right-hand panel of Figure 2A)



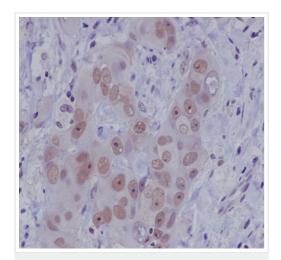
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM antibody [Y170] (ab32420)

Formaldehyde-fixed human colon tissue stained for ATM using ab32420 at 1/100 dilution in immunohistochemical analysis.



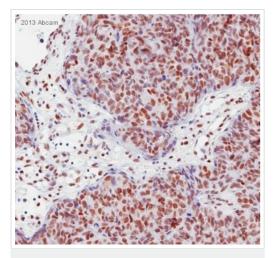
Flow Cytometry (Intracellular) - Anti-ATM antibody [Y170] (ab32420)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labelling ATM with purified ab32420 at 1/20 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. An Alexa Fluorr®488-conjugated goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



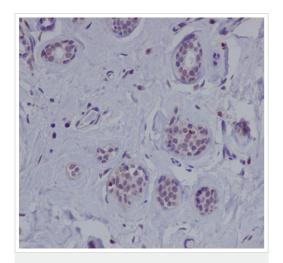
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM antibody [Y170] (ab32420)

Immunohistochemical analysis of paraffin-embedded Human breast cancer tissue labeling ATM with ab32420 at 1:100 dilution. Tissue underwent antigen retrieval using Tris/EDTA Buffer (pH9.0). The section was counterstained with haematoxylin.



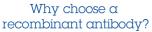
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM antibody [Y170] (ab32420)

Formaldehyde-fixed human serous ovarian tumor tissue stained for ATM using ab32420 at 1/50 dilution in immunohistochemical analysis.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM antibody [Y170] (ab32420)

Immunohistochemical analysis of paraffin-embedded Human normal breast tissue labeling ATM with ab32420 at 1:100 dilution. Tissue underwent antigen retrieval using Tris/EDTA Buffer (pH9.0). The section was counterstained with haematoxylin.





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Consistent and reproducible results



Success from the first experiment
Confirmed specificity



Ethical standards compliant Animal-free production

Anti-ATM antibody [Y170] (ab32420)

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