

Product datasheet

Anti-ATM (phospho S1981) antibody [10H11.E12] ab36810

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Overview

Product name	Anti-ATM (phospho S1981) antibody [10H11.E12]
Description	Mouse monoclonal [10H11.E12] to ATM (phospho S1981)
Specificity	ab36810 is specific for the human ATM kinase.
Tested applications	Suitable for: Flow Cyt, WB, ICC, IHC-P
Species reactivity	Reacts with: Rat, Human
Immunogen	Synthetic peptide (Human) with a phosphorylated serine (1981)
Positive control	Irradiated normal Human fibroblasts (no reactivity against non-irradiated cell extracts).
General notes	This antibody clone is manufactured by Abcam.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.
Storage buffer	pH: 7.4 Preservative: 0.05% Sodium azide Constituent: PBS
	Contains 0.4M Arginine
Purity	IgG fraction
Clonality	Monoclonal
Clone number	10H11.E12
Isotype	IgG1
Light chain type	kappa

Applications

Our [Abpromise guarantee](#) covers the use of **ab36810** 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		Use 1µg for 10 ⁶ cells. ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.
WB		1/1000. Detects a band of approximately 370 kDa (predicted molecular weight: 370 kDa). Abcam recommends using 3% Milk as the blocking agent.
ICC		1/1000.
IHC-P	★★★★★	Use a concentration of 4 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Target

Function

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.

Tissue specificity

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

Involvement in disease

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.

Sequence similarities

Belongs to the PI3/PI4-kinase family. ATM subfamily.

Contains 1 FAT domain.

Contains 1 FATC domain.

Contains 1 PI3K/PI4K domain.

Domain

The FATC domain is required for interaction with KAT5.

Post-translational modifications

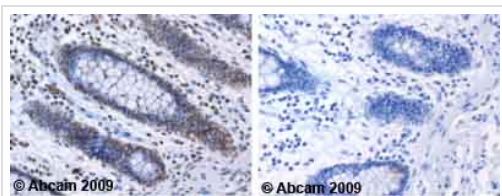
Phosphorylated by NUAK1/ARK5. Autophosphorylation on Ser-367, Ser-1893, Ser-1981 correlates with DNA damage-mediated activation of the kinase.

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.

Cellular localization

Nucleus. Cytoplasmic vesicle. Primarily nuclear. Found also in endocytic vesicles in association with beta-adaptin.

Images



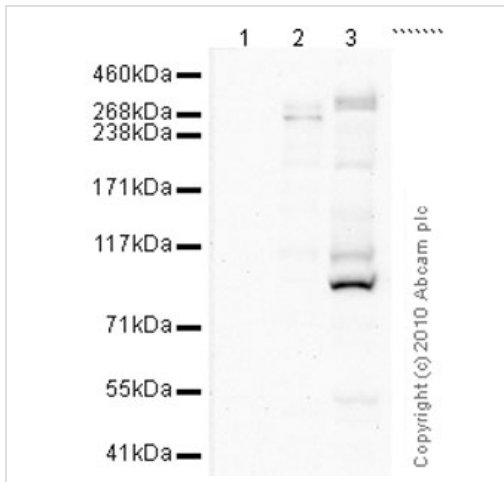
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATM (phospho S1981) antibody [10H11.E12] (ab36810)

Ab36810 staining Human colon. Staining is localised to the nucleus.

Left panel: with primary antibody at 4 ug/ml.

Right panel: isotype control.

Sections were stained using an automated system DAKO Autostainer Plus , at room temperature. Sections were rehydrated and antigen retrieved with the DAKO 3-in-1 antigen retrieval buffer citrate pH 6.0 in a DAKO PT Link. Slides were peroxidase blocked in 3% H₂O₂ in methanol for 10 minutes. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 minutes and detected with Dako Envision Flex amplification kit for 30 minutes. Colorimetric detection was completed with diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that for manual staining we recommend to optimize the primary antibody concentration and incubation time (overnight incubation), and amplification may be required.



Western blot - Anti-ATM (phospho S1981) antibody [10H11.E12] (ab36810)

All lanes : Anti-ATM (phospho S1981) antibody [10H11.E12] (ab36810) at 10 µg/ml

Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate

Lane 2 : Extract from Patient with Ataxia-Telangiectasia Whole Cell Lysate

Lane 3 : Irradiated HeLa Whole Cell Lysate

Lysates/proteins at 20 µg per lane.

Secondary

Goat polyclonal to Mouse IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution
Developed using the ECL technique

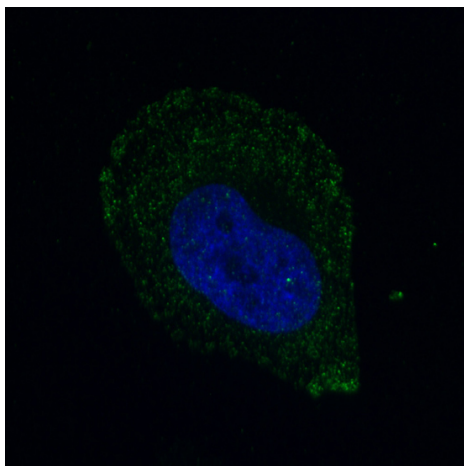
Performed under non-reducing conditions.

Predicted band size : 370 kDa

Observed band size : 370 kDa

Additional bands at : 100 kDa, 110 kDa, 145 kDa, 200 kDa. We are unsure as to the identity of these extra bands.

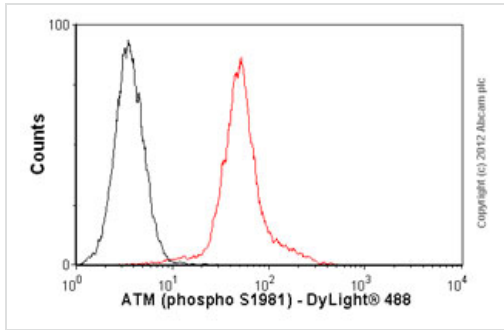
Exposure time : 20 minutes



Immunocytochemistry/ Immunofluorescence - Anti-ATM (phospho S1981) antibody [10H11.E12] (ab36810)

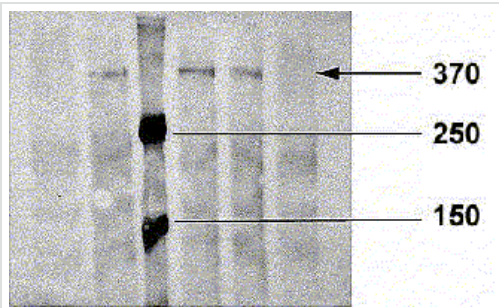
This image is courtesy of an Abreview submitted by Dimitra Kalamida

ab36810 staining ATM (phospho S1981) in T98G Human brain glioblastoma cell by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde, permeabilized with 0.1% Triton X-100, pH 7.4 for 5 minutes at room temperature and blocked with 5% BSA for 20 minutes at room temperature. Samples were incubated with primary antibody (1/250 in PBS) for 1 hour. A CF488A-conjugated Goat anti-mouse IgG polyclonal (1/500) was used as the secondary antibody.



Flow Cytometry - Anti-ATM (phospho S1981) antibody [10H11.E12] (ab36810)

Overlay histogram showing HeLa cells stained with ab36810 (red line). The cells were fixed with 80% methanol (5 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 (ab36810, 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. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Western blot - Anti-ATM (phospho S1981) antibody [10H11.E12] (ab36810)

All lanes : Anti-ATM (phospho S1981) antibody [10H11.E12] (ab36810) at 1/1000 dilution

Lane 1 : Control lane

Lane 2 : Irradiated Human fibroblasts (10 Gy gamma-irradiation)

Lane 3 : Molecular weight marker

Lane 4 : Peroxidated Human fibroblasts (300 µM hydrogen peroxide)

Lane 5 : Peroxidated Human fibroblasts (1 mM hydrogen peroxide)

Lane 6 : Peroxidated Human fibroblasts (10 mM hydrogen peroxide)

Developed using the ECL technique

Predicted band size : 370 kDa

Observed band size : 370 kDa

Lysate loading concentration: 40µg

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