abcam

Product datasheet

Anti-ATM (phospho S1981) antibody [EP1890Y] ab81292

Recombinant RabMAb

★★★★★ 18 Abreviews 298 References 11 Images

Overview

Product name Anti-ATM (phospho S1981) antibody [EP1890Y]

Description Rabbit monoclonal [EP1890Y] to ATM (phospho S1981)

Host species Rahhit

Tested applications Suitable for: Dot blot, Flow Cyt (Intra), WB, IHC-P, IP

Unsuitable for: ICC/IF

Species reactivity Reacts with: Human

Immunogen Synthetic peptide within Human ATM (phospho S1981). The exact sequence is proprietary.

(Peptide available as ab235690)

Positive control WB: HEK-293 cell lysate treated with doxorubicin. IHC-P: Human gastric carcinoma, breast

> carcinoma, tonsil, cervical carcinoma, hepatocellular carcinoma and endometrial carcinoma tissues and mouse endometrium tissue. IP: HEK-293 cell lysate treated with doxorubicin. Flow

Cyt (intra): HepG2 cells

General notes 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.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

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

species. Please contact us for more information.

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.

Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EP1890Y

Isotype IgG

Applications

The Abpromise quarantee

Our Abpromise guarantee covers the use of ab81292 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 |
|------------------|-------------------|--|
| Dot blot | | 1/100000. |
| Flow Cyt (Intra) | | 1/60. |
| WB | ★★★★★ (11) | 1/50000. Detects a band of approximately 370 kDa (predicted molecular weight: 351 kDa). We highly recommend using a positive control such as HEK293 cells treated with 10uM doxorubicin for 24 hours. |
| IHC-P | ★★★★★ (2) | 1/70 - 1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols. |
| IP | | 1/30 - 1/40. |

Application notes

Is unsuitable for ICC/IF.

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.

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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.

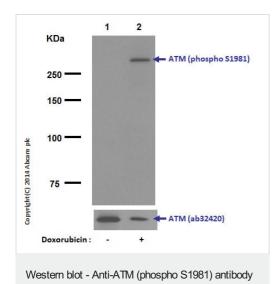
Cellular localization

[EP1890Y] (ab81292)

 $\hbox{\it Nucleus. Cytoplasmic vesicle. Primarily nuclear. Found also in endocytic vesicles in association}$

with beta-adaptin.

Images



All lanes : Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292) at 1/50000 dilution (purified)

Lane 1: HEK-293 (Human epithelial cell line from embryonic

kidney) cell lysate, untreated

Lane 2: HEK-293 cell lysate, treated with Doxorubicin

Lysates/proteins at 10 µg per lane.

Secondary

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

Predicted band size: 351 kDa Observed band size: 370 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

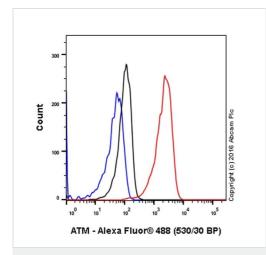
Secondary-antibody

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human hepatocellular carcinoma tissue labelling ATM (phospho S1981) with purified ab81292 at 1/70. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. ab97051, an HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500).

Negative control using PBS instead of primary antibody.

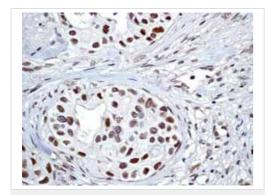
Counterstained with hematoxylin.



Flow Cytometry (Intracellular) - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

Intracellular Flow Cytometry analysis of HepG2 (Human hepatocellular carcinoma) cells labeling ATM (phospho S1981) with purified ab81292 at 1/60 dilution (10µg/mL) (red).

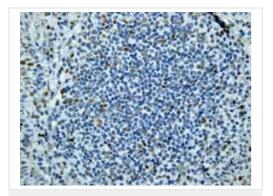
Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A Goat anti rabbit IgG (Alexa Fluorr[®]488) at 1/2000 dilution was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) was used as the unlabeled control.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast carcinoma tissue labeling ATM with unpurified ab81292.

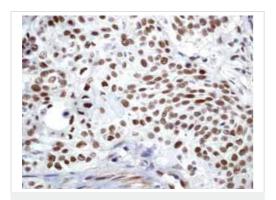
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of normal human tonsil tissue labeling ATM with unpurified ab81292.

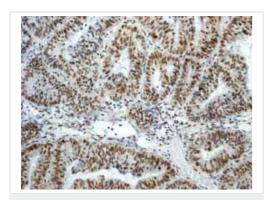
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervical carcinoma tissue labeling ATM with unpurified ab81292.

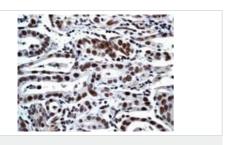
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human endometrial carcinoma tissue labeling ATM with unpurified ab81292.

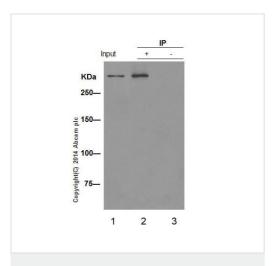
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human gastric carcinoma tissue labeling ATM with unpurified ab81292 at a 1/100 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-ATM (phospho S1981) antibody [EP1890Y] (ab81292)

ATM was immunoprecipitated from HEK-293 (Human embryonic kidney epithelial cell) treated with Doxorubicin whole cell lysate with ab81292 at 1/30 dilution (5 μ g in 1 mg lysates). Western blot was performed from the immunoprecipitate using ab81292 at 1/2000 dilution. An anti-Rabbit lgG (HRP), specific to the non-reduced form of lgG was used as secondary antibody at 1/1500 dilution.

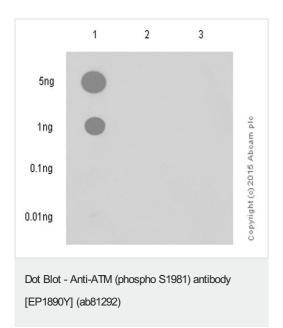
Lane 1: HEK-293 treated with Doxorubicin whole cell lysate 10 μ g (Input).

Lane 2: ab81292 IP in HEK-293 treated with Doxorubicin whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab81292 in HEK-293 treated with Doxorubicin whole cell lysate.

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.



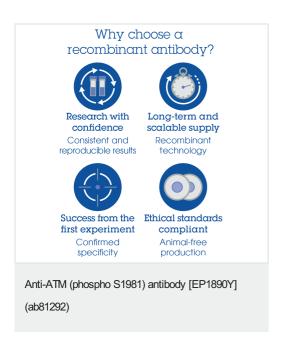
Dot blot analysis of ATM peptides using ab81292 at 1/000 dilution followed by Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated secondary antibody (ab97051) at 1/100,000 dilution.

Blocking and diluting buffer was 5% NFDM/TBST, exposure time 3 minutes.

Lane 1: ATM (pS1981) phospho peptide

Lane 2: ATM non-phospho peptide

Lane 3: ATM (pS428) phospho peptide



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