

# Human ATM knockout A549 cell line ab276095

4 Images

### Overview

Product name	Human ATM knockout A549 cell line
Parental Cell Line	A549
Organism	Human
Passage number	<20
Tested applications	<b>Suitable for:</b> WB
Biosafety level	1
General notes	<p><b>Recommended control:</b> Human wild-type A549 cell line (<a href="#">ab275463</a>). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1.</p> <p><b>Cryopreservation cell medium:</b> Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p><b>Culture medium:</b> F-12K + 10% FBS</p> <p><b>Initial handling guidelines:</b> Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability.</p> <ol style="list-style-type: none"> <li>1. Thaw the vial in 37°C water bath for approximately 1-2 minutes.</li> <li>2. Transfer the cell suspension (0.8 mL) to a 15 mL/50 mL conical sterile polypropylene centrifuge tube containing 8.4 mL pre-warmed culture medium, wash vial with an additional 0.8 mL culture medium (total volume 10 mL) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 mL represents minimum recommended dilution. 20 mL represents maximum recommended dilution.</li> <li>3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of <math>2 \times 10^3</math>-<math>1 \times 10^4</math> cells/cm<sup>2</sup>. Seeding density is given as a guide only and should be scaled to align with individual lab schedules.</li> <li>4. Incubate the culture at 37°C incubator with 5% CO<sub>2</sub>. Cultures should be monitored daily.</li> </ol> <p><b>Subculture guidelines:</b></p> <p>All seeding densities should be based on cell counts gained by established methods. A guide seeding density of <math>6 \times 10^4</math> cells/cm<sup>2</sup> is recommended.</p> <p>A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required.</p> <p>Cells should be passaged when they have achieved 80-90% confluence.</p> <p>Do not exceed <math>7 \times 10^4</math> cells/cm<sup>2</sup>.</p>

This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our [limited use license](#) and [patent pages](#).

We will provide viable cells that proliferate on revival.

## Properties

<b>Number of cells</b>	1 x 10 <sup>6</sup> cells/vial, 1 mL
<b>Adherent /Suspension</b>	Adherent
<b>Tissue</b>	Lung
<b>Cell type</b>	epithelial
<b>Disease</b>	Carcinoma
<b>Gender</b>	Male
<b>Mycoplasma free</b>	Yes
<b>Storage instructions</b>	Shipped on Dry Ice. Store in liquid nitrogen.
<b>Storage buffer</b>	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

## Target

<b>Function</b>	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.
<b>Tissue specificity</b>	Found in pancreas, kidney, skeletal muscle, liver, lung, placenta, brain, heart, spleen, thymus, testis, ovary, small intestine, colon and leukocytes.
<b>Involvement in disease</b>	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-polymphocytic 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 NUA1/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.

## Applications

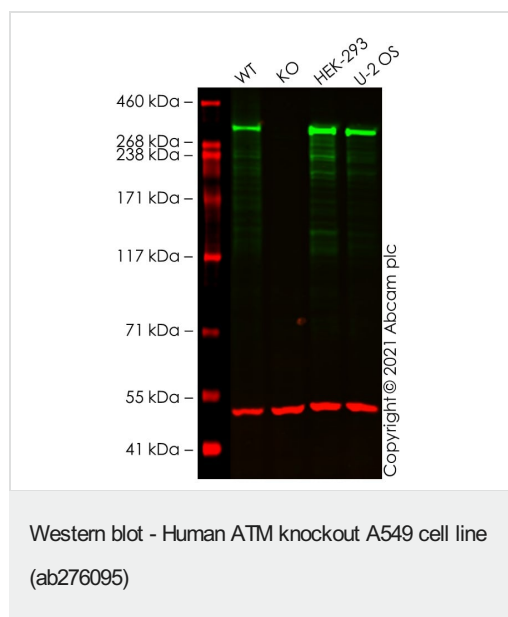
### The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab276095 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
WB		Use at an assay dependent concentration.

## Images



**All lanes** : Anti-ATM antibody [EPR20100] - ChIP Grade (**ab201022**) at 1/1000 dilution

**Lane 1** : Wild-type A549 cell lysate

**Lane 2** : ATM knockout A549 cell lysate

**Lane 3** : HEK-293 cell lysate

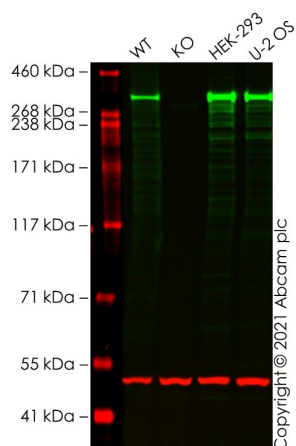
**Lane 4** : U-2 OS cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Observed band size:** 350 kDa

False colour image of Western blot: Anti-ATM antibody [EPR20100] - ChIP Grade 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, [ab201022](#) 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 IgG H&L (IRDye® 680RD) preabsorbed ([ab216776](#)) at 1/20000 dilution.



Western blot - Human ATM knockout A549 cell line (ab276095)

**All lanes :** Anti-ATM antibody [EPR17059] ([ab199726](#)) at 1/2000 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

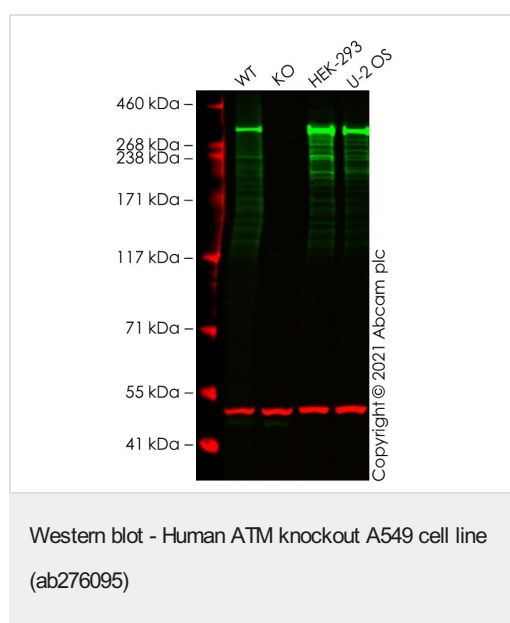
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Observed band size:** 350 kDa

False colour image of Western blot: Anti-ATM antibody [EPR17059] staining at 1/2000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] ([ab7291](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, [ab199726](#) 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 IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) 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

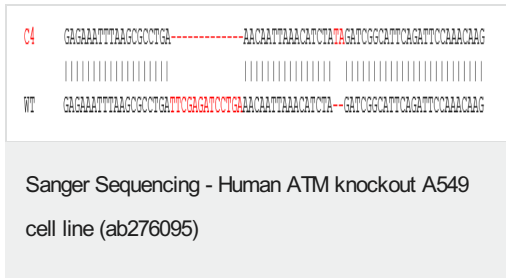
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**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 IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) at 1/20000

dilution.



Knockout achieved by using CRISPR/Cas9, Homozygous: 28 bp deletion in exon 3

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

### Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <https://www.abcam.com/abpromise> or contact our technical team.

### Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors