

## Anti-ATM Protein Kinase pS1981 (MOUSE) Monoclonal Antibody - 200-301-400

**Code:** 200-301-400

**Size:** 100 µg

**Product Description:** Anti-ATM Protein Kinase pS1981 (MOUSE) Monoclonal Antibody - 200-301-400

**Concentration:** 1.0 mg/mL by UV absorbance at 280 nm

**PhysicalState:** Liquid (sterile filtered)

<b>Label</b>	Unconjugated
<b>Host</b>	Mouse
<b>Gene Name</b>	ATM
<b>Species Reactivity</b>	human, mouse, rat
<b>Buffer</b>	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
<b>Stabilizer</b>	None
<b>Preservative</b>	0.01% (w/v) Sodium Azide
<b>Storage Condition</b>	Store Anti-ATM phospho S1981 Antibody at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
<b>Synonyms</b>	mouse anti-ATM antibody, mouse anti-ATMpS1981 antibody, mouse anti- ATM pS1981 antibody, DKFZp781A0353 antibody, Human phosphatidylinositol 3 kinase homolog antibody, MGC74674 antibody, Serine protein kinase ATM antibody, T cell prolymphocytic leukemia antibody
<b>Application Note</b>	Protein A Purified Mab anti-ATM has been tested by ELISA, Flow Cytometry, IF, and western blotting against both the native and recombinant forms of the protein. The antibody immunoprecipitates ATM from irradiated human and transfected mouse cells. By immunofluorescence, foci are detected in irradiated human and mouse fibroblasts. This antibody is not recommended for immunohistochemistry. Instead, for IHC, use the clone 7C10D8 (p/n 200-301-500).
<b>Background</b>	Anti ATM pS1981 Antibody is a phospho site specific antibody and recognizes the product of the ATM gene that is mutated in the hereditary disease ataxia-telangiectasia. ATM codes for a protein kinase that acts as a master regulator of cellular responses to DNA double-strand breaks. ATM is normally inactive and the question of how it is activated in the event of DNA damage (due to ionizing radiation for instance) is central to understanding its function. ATM protein is now shown to be present in undamaged cells as an inactive dimer. Low doses of ionizing radiation, which induce only a few DNA breaks, activate at least half of the total ATM protein present, possibly in response to changes in chromatin structure. The ATM gene encodes a 370-kDa protein that belongs to the phosphoinositide 3-kinase (PI(3)K) superfamily, but which phosphorylates proteins rather than lipids. The 350-amino-acid kinase domain at the carboxy terminus of this large protein is the only segment of ATM with an assigned function. Exposure of cells to IR triggers ATM kinase activity, and this function is required for arrests in G1, S and G2 phases of the cell cycle. Several substrates of the ATM kinase participate in these IR-induced cell-cycle arrests. These include p53, Mdm2 and Chk2 in the G1 checkpoint; Nbs1, Brca1, FancD2 and SMC1 in the transient IR-induced S-phase arrest; and Brca1 and hRad17 in the G2/M checkpoint. Ideal for Cancer, Cell Signaling, Chromatin, Neuroscience and Signal Transduction research.
<b>Purity And Specificity</b>	Anti-ATM phospho S1981 Monoclonal Antibody is directed against human ATM and is useful in determining its presence in various assays. This monoclonal anti-ATM antibody recognizes the phosphorylated epitope in native and over-expressed proteins found in various tissues and extracts. By ELISA reactivity against SLAFEEGSpQSTTISS at a 1:1600 dilution shows an absorbance >3.000; whereas reactivity against SLAFEEGSpQSTTISS shows an absorbance of 0.145. Reactivity is observed against human ATM. Cross reactivity with ATM from other mammalian sources has not been tested. The immunogen has 91% sequence homology with mouse ATM.
<b>Assay Dilutions</b>	User Optimized
<b>ELISA</b>	1:20,000 - 1:100,000
<b>Western Blot</b>	1:200 - 1:2,000
<b>Immunohistochemistry</b>	Not Recommended
<b>IF Microscopy</b>	1:100 - 1:500
<b>Flow Cytometry</b>	5 µg/mL
<b>Other Assays</b>	User Optimized
<b>Expiration</b>	Expiration date is one (1) year from date of opening.

## Immunogen

Anti-ATM phospho S1981 Antibody was produced from a synthetic peptide S-L-A-F-E-E-G-Sp-Q-S-T-T-I-S-S corresponding to aa 1974-1988 of human ATM.

## Specific Reference

Bakkenist, C. J. & Kastan, M. B. (2003). DNA damage activates ATM through intermolecular autophosphorylation and dimer dissociation. *Nature* 421, 499-506. Kitagawa R, Bakkenist CJ, McKinnon PJ, Kastan MB. (2004) Phosphorylation of SMC1 is a critical downstream event in the ATM-NBS1-BRCA1 pathway. *Genes Dev.* 18(12):1423-38. Falck, J. Coates, J. and Jackson, S.P. (2005) Conserved modes of recruitment of ATM, ATR and DNA-PKcs to sites of DNA damage. *Nature* 434: 605-611. Bartkova J, Horejsi Z, Koed K, Kramer A, Tort F, Zieger K, Guldborg P, Sehested M, Nesland JM, Lukas C, Orntoft T, Lukas J, Bartek J. (2005) DNA damage response as a candidate anti-cancer barrier in early human tumorigenesis. *Nature* 434: 864-870. Bartkova J, Bakkenist CJ, Rajpert-De Meyts E, Skakkebaek NE, Sehested M, Lukas J, Kastan MB, Bartek J. (2005) ATM Activation in Normal Human Tissues and Testicular Cancer. *Cell Cycle* 4;(6) [Epub ahead of print]. Pusapati, R. V., Rounbehler, R. J., Hong, S., Powers, J. T., Yan, M., Kiguchi, K., ... & Johnson, D. G. (2006). ATM promotes apoptosis and suppresses tumorigenesis in response to Myc. *Proceedings of the National Academy of Sciences of the United States of America*, 103(5), 1446-1451. Zhang, W., Peng, G., Lin, S. Y., & Zhang, P. (2011). DNA damage response is suppressed by the high cyclin-dependent kinase 1 activity in mitotic mammalian cells. *Journal of Biological Chemistry*, 286(41), 35899-35905.

## Related Products

000-000-400	ATM pS1981 CONTROL PEPTIDE - 000-000-400
100-401-264	Anti-NFKB p65 (Rel A) pS276 (RABBIT) Antibody - 100-401-264
200-301-397	Anti-SMC1 pS957 (MOUSE) Monoclonal Antibody - 200-301-397
600-401-398	Anti-ATM Protein Kinase S1981 (RABBIT) Antibody - 600-401-398

## Related Links

UniProtKB - Q13315

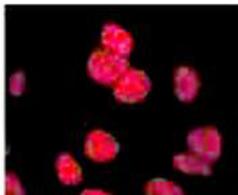
<http://www.uniprot.org/uniprot/Q13315>

NCBI - Q13315.3 <http://www.ncbi.nlm.nih.gov/protein/Q13315.3>

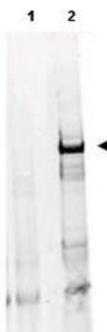
GenElD - 472

## Images

- 1 Anti ATM antibody showing overlay of anti-ATM pS1981 staining. Cells were fixed 15 min after 5 Gy (IR+) of irradiation, then labeled with antibody. See Kitagawa et al. for additional details.

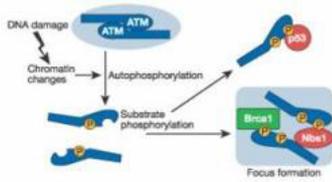


- 2 Anti ATM Mab with human derived HEK293 cells treated with doxorubicin using Rockland's Protein A Purified Mab anti-ATM Protein Kinase pS1981(clone 10H11.E12). A 370 kDa band corresponding to phosphorylated ATM is detected (arrowhead, lane 2). The lysate was prepared with HALT phosphatase inhibitor (Pierce). Pre-incubation of antibody with immunizing phospho peptide negates specific staining (lane 1). Approximately 30 µg of lysate was added to each lane of an SDS-PAGE gel under non-reducing conditions. The protein was transferred to nitrocellulose using standard methods. After blocking the membrane was probed with the primary antibody diluted 1:500 overnight at 4°C followed by washes and reaction with a 1:10,000 dilution of IRDye™800 conjugated Gt-a-Mouse IgG [H&L] (code 610-132-121) for 40 min at room temperature. LICOR's Odyssey® Infrared Imaging System was used to scan and process the image. Other detection systems will yield similar results.



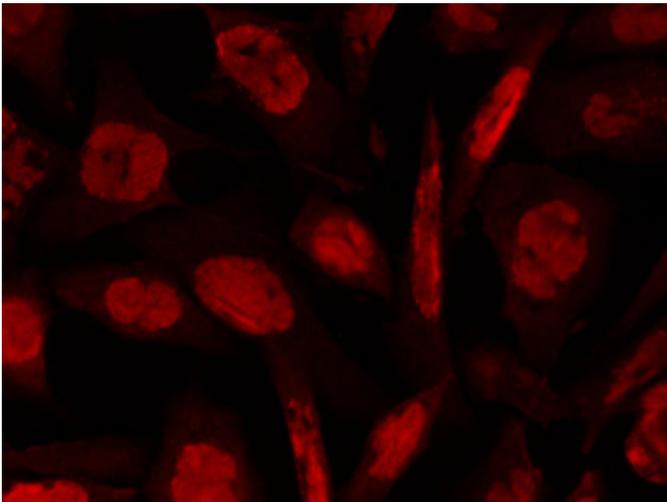
3

## Rockland Mouse Anti-ATM Protein Kinase pS1981 Antibody.



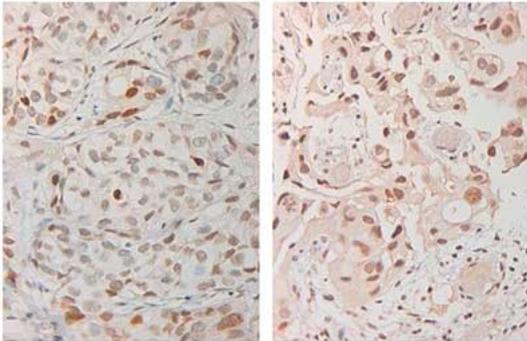
4

Rockland's anti-ATM pS1981 mouse monoclonal antibody (Catalog # 200-301-400) detects ATM phosphorylated on Ser 1981 by Indirect immunofluorescence microscopy. Shown are hTCEpi cells (courtesy of Dr. Danielle Robertson) infected with HSV-1 at MOI 5.0 and fixed at 8 hpi with 3% paraformaldehyde/2% sucrose for 10 min. After rinsing, cells were permeabilized with 0.5% Triton X-100 for 5 min, blocked with 3% BSA for 30 min, and stained with Rockland's primary anti-ATM pS1981 antibody overnight at 5  $\mu\text{g}/\text{mL}$  (1:200). Secondary staining was performed with Alexa Fluor 594 anti-mouse antibody. Images were taken with Olympus AX70 compound epifluorescence microscope equipped with Spot RT Slider camera. Experiment was performed by Oleg Alekseev in the laboratory of Dr. Jane Azizkhan-Clifford at Drexel University College of Medicine.



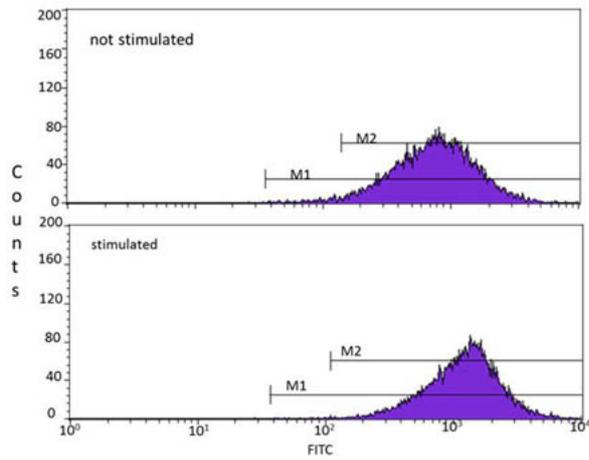
5

Immunohistochemistry with anti-ATM Antibody. Tissue: Human Bladder Cancer. Fixation: FFPE buffered formalin 10% conc. Ag Retrieval: HIER citrate buffer pH6 (left) or HIER EDTA pH9 (Right). Primary antibody: anti-ATM at 2  $\mu\text{g}/\text{ml}$  for 2 hr. Secondary Ab: anti-rabbit polymer HRP for 20min at RT.



6

Flow Cytometry of Mouse anti-ATMpS1981 antibody. Cells: HEK293. Stimulation: none – top image, 0.1mg/ml Zeocin for 3 hr – bottom image. Primary antibody: anti-ATM pS1981 antibody at 5  $\mu\text{g}/\text{mL}$  for 30 min at 4°C. Secondary antibody: anti-mouse IgG FITC at 1 $\mu\text{g}/\text{ml}$ , 30min at 4°C IN THE DARK.



### Disclaimer

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