

**Certificate of Analysis**
**Product:** Protein A Purified Mouse Mab anti-ATM Protein Kinase pS1981

**Code:** 200-301-500

**Lot #:** 20773

**Size:** 100 µg

**Physical State:** Liquid (sterile filtered)

**Antibody Concentration:** 1.0 mg/ml (by UV absorbance at 280 nm)

**Buffer:** 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

**Stabilizer:** None

**Preservative:** 0.01% (w/v) Sodium Azide

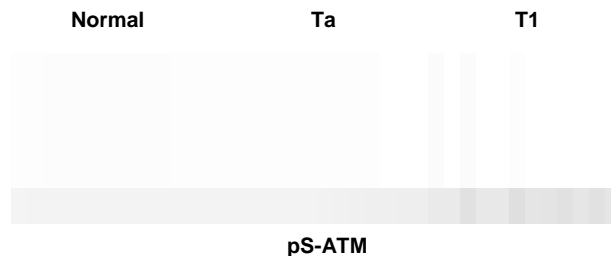
**Clone:** 7C10D8 (IgG<sub>2aκ</sub>)

**Fusion Partner:** Sp2/0 mouse myeloma

**Storage Conditions:** Store vial at -20° C. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Dilute only prior to immediate use. Expiration date is one (1) year from date of restoration.

**Background Information:** *ATM*, the gene mutated in the hereditary disease ataxia-telangiectasia, 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 G<sub>1</sub>, S and G<sub>2</sub> 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 G<sub>1</sub> checkpoint; Nbs1, Brca1, FancD2 and SMC1 in the transient IR-induced S-phase arrest; and Brca1 and hRad17 in the G<sub>2</sub>/M checkpoint.

**Figure 1.** Immunohistochemistry was used to show constitutive activation of the ATM pathway in human urinary bladder cancer. Anti-ATM Protein Kinase pS1981 (clone # 7C10D8 ) was used to stain normal uroepithelium, early superficial lesions (Ta), and earliest invasive (T1) primary carcinomas. ATM protein is ubiquitously expressed, but Ser 1981-phosphorylated ATM (pS-ATM) is detectable only in tumor tissues. See Bartkova et al. for additional details. Reprinted by permission from Nature (*Cell Cycle* 4;(6), 2005) Macmillan Publishers Ltd.


**Recommended Dilutions:**

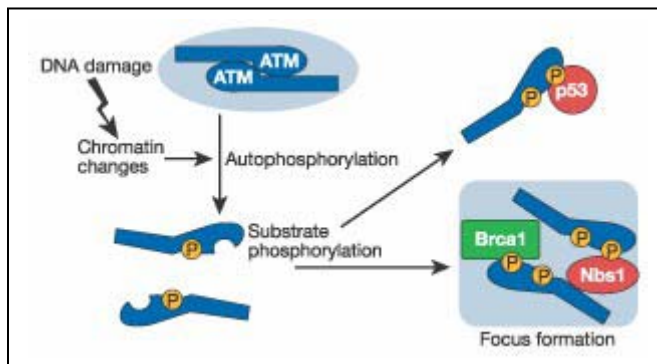
<b>ELISA</b>	User Optimized
<b>WESTERN BLOT</b>	User Optimized
<b>IF MICROSCOPY</b>	User Optimized
<b>IMMUNOHISTOCHEMISTRY</b>	1:100 - 1:500
<b>OTHER APPLICATIONS</b>	User Optimized

**Application Note(s):** This antibody clone has been optimized for IHC, but may also be used for western blotting, immunoprecipitation or immunofluorescence microscopy. Indirect immunoperoxidase staining on formaldehyde-fixed, de-paraffinized tissue sections and/or formalin-fixed cultured cells are recommended when using this antibody as

described in Bartkova et al 2005. Product p/n 200-301-400 produced from clone 10H11.E12 has been optimized for WB, IP and IF.

**Purity and Specificity:** This Protein A Purified Mab antibody is directed against human ATM and is useful in determining its presence by immunohistochemistry. This monoclonal anti-ATM antibody recognizes the phosphorylated form of the protein in native and over-expressed proteins found in various tissues and extracts. Reactivity is observed against human and mouse ATM. Cross reactivity with ATM from other mammalian sources has not been tested.

**Immunogen:** This 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.



#### Related Product(s):

#600-401-398	Affinity Purified anti-ATM Protein Kinase S1981 [Rabbit]
#200-301-400	Protein A Purified Mouse Mab anti-ATM Protein Kinase pS1981 for WB, IF and IP
#200-301-500	Protein A Purified Mouse Mab anti-ATM Protein Kinase pS1981 for IHC
#200-302-400	Fluorescein Conjugated Protein A Purified Mouse Mab anti-ATM Protein Kinase pS1981
#200-306-400	Biotin Conjugated Protein A Purified Mouse Mab anti-ATM Protein Kinase pS1981
#200-303-400	Peroxidase Conjugated Protein A Purified Mouse Mab anti-ATM Protein Kinase pS1981
#000-000-398	CONTROL PEPTIDE for 600-401-398 anti-ATM Protein Kinase S1981
#000-000-400	CONTROL PEPTIDE for 600-401-400 anti-ATM Protein Kinase pS1981
#611-703-127	HRP Anti-Rabbit IgG [H&L] MX10 (DONKEY)
#611-132-122	IRDye800 Anti-Rabbit IgG [H&L] MX10 (GOAT)
#W09-000-360	Human Derived MCF-7 Whole Cell Lysate (Ready-to-Use)
#W09-000-366	Hydrogen Peroxide Stimulated Human Derived MCF-7 Whole Cell Lysate (Ready-to-Use)

#### Specific References:

[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].

**Note:** This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 326, Gilbertsville, Pennsylvania, USA. This antibody and certain aspects of its use are disclosed and claimed in pending U.S. Patent Applications published as U.S. Patent Publication Nos. 2003/0077661 and 2003/0157572.