ROCKLAND
Immunochemicals for Research

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Product Specification Sheet

Product: Fluorescein Conjugated Protein A Purified Mab anti-ATM Protein Kinase pS1981 [Mouse]

Code: 200-302-400 **Lot #:** 13196

Size: 100 μg **Physical State:** Lyophilized

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

Label: Fluorescein isothiocyanate (FITC) (Molecular Weight 390 daltons)

Absorption Wavelength: 495 nm **Emission Wavelength**: 528 nm

Fluorochrome/Protein Ratio: 3.8 moles FITC per mole of Mouse IgG

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

Stabilizer: 10 mg/ml Bovine Serum Albumin (BSA) IgG and Protease free

Preservative: 0.01% (w/v) Sodium Azide

Clone: 10H11.E12 ($IgG_1\kappa$) **Fusion Partner:** Sp2/0 mouse myeloma

Storage Conditions: Store vial at 4° C prior to restoration. Restore with 0.1 ml of deionized water (or equivalent). For extended storage mix product with glycerol to 50% and then aliquot contents and freeze at -20° C or below. 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. 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_1, S and G_2 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_1 checkpoint; Nbs1,

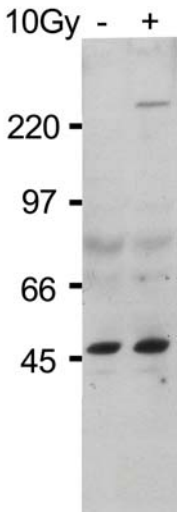
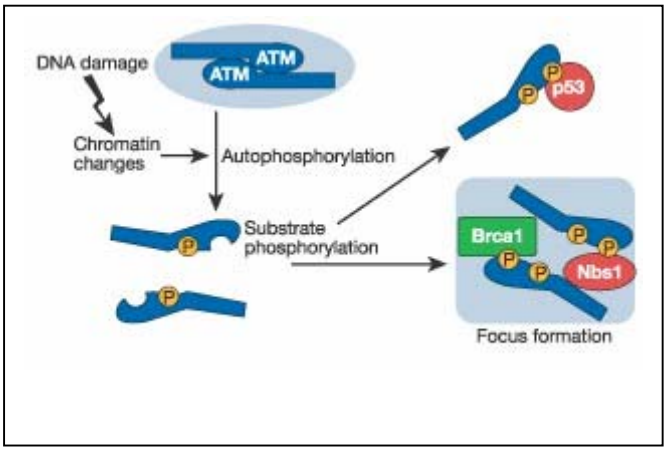


Figure1. Anti-ATM pS1981 polyclonal antibody detects ATM in human fibroblast cell lysate after exposure to ionizing radiation. The (-) shows a lysate from a similarly prepared mock irradiation. The apparent molecular weight of ATM is 370 kDa. The induction of ATM in fibroblasts is consistent with experiments described in Bakkenist and Kastan. After blocking the membrane was probed with a 1:600 dilution of the primary antibody for 1 h at room temperature followed by washes and reaction with a 1:2000 dilution of HRP conjugated Dnky-a-Rabbit IgG [H&L] (code 611-703-127) for 30 min at room temperature. Other detection systems will yield similar results.

Brca1, FancD2 and SMC1 in the transient IR-induced S-phase arrest; and Brca1 and hRad17 in the G_2/M checkpoint. See Bakkenist, C. J. & Kastan, M. B. *Nature* **421**, 499-506 (2003) for a complete presentation of this antibody's specificity and utility.

Application Note(s): This product is suitable for immunomicroscopy and flow cytometry or FACS analysis as well as other antibody based fluorescent assays requiring lot-to-lot consistency.

Recommended Dilutions: A 1:500 dilution is recommended for most fluorescent immunochemical assays. Generally a 1:50 to 1:100 dilution is suitable for flow cytometry/FACS. Optimal dilutions should be determined by the researcher for other applications.



Purity and Specificity: This Protein A Purified Mab 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. 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]
#600-401-400	Affinity Purified anti-ATM Protein Kinase pS1981 [Rabbit]
#200-301-400	Protein A Purified Mab anti-ATM Protein Kinase pS1981 [Mouse]
#200-302-400	Fluorescein Conjugated Protein A Purified Mab anti-ATM Protein Kinase pS1981 [Mouse]
#200-306-400	Biotin Conjugated Protein A Purified Mab anti-ATM Protein Kinase pS1981 [Mouse]
#200-303-400	Peroxidase Conjugated Protein A Purified Mab anti-ATM Protein Kinase pS1981 [Mouse]
#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
#600-401-383	Affinity Purified anti-FLAG EPITOPE TAG (Rabbit)
#200-301-174	Protein A Purified Mouse Monoclonal Anti-Human p53
#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)

General References:

None yet for this monoclonal, but results from use of a polyclonal antibody with similar specificity were reported in: Bakkenist, C. J. & Kastan, M. B. (2003). DNA damage activates ATM through intermolecular autophosphorylation and dimer dissociation. *Nature* **421**, 499-506.

see also related commentary, Bartek, J. and Lukas, J., *Nature 421:* 486-488 (2003).

Conjugation Reference: The and Feltkamp, *Immunology* **18**; 865, 1970.

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.