

Product datasheet

Anti-ATR (phospho S428) antibody [EPR2184] ab178407

Recombinant RabMAb

15 References 5 Images

Overview

Product name	Anti-ATR (phospho S428) antibody [EPR2184]
Description	Rabbit monoclonal [EPR2184] to ATR (phospho S428)
Host species	Rabbit
Specificity	<i>Stimulation may be required to allow detection of the phosphorylated protein. Please see images below for recommended treatment conditions and positive controls.</i>
Tested applications	Suitable for: Dot blot, IHC-P, WB Unsuitable for: Flow Cyt, ICC/IF or IP
Species reactivity	Reacts with: Human Does not react with: Mouse, Rat
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	IHC: Human breast tissue, human thyroid tissue; WB: Untreated HeLa, HeLa treated with 4mM hydroxyurea for 20 hours whole cell lysate;
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p> <p>We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.</p>

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 long

term. Avoid freeze / thaw cycle.

Storage buffer

pH: 7.20
Preservative: 0.01% Sodium azide
Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.5% BSA

Purity

Protein A purified

Clonality

Monoclonal

Clone number

EPR2184

Isotype

IgG

Applications

The Abpromise guarantee

Our [Abpromise guarantee](#) covers the use of ab178407 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		1/1000.
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
WB		1/1000 - 1/10000. Predicted molecular weight: 300 kDa.

Application notes

Is unsuitable for Flow Cyt, ICC/IF or IP.

Target

Function

Serine/threonine protein kinase which activates checkpoint signaling upon genotoxic stresses such as ionizing radiation (IR), ultraviolet light (UV), or DNA replication stalling, thereby acting as a DNA damage sensor. Recognizes the substrate consensus sequence [ST]-Q. Phosphorylates BRCA1, CHEK1, MCM2, RAD17, RPA2, SMC1 and p53/TP53, which collectively inhibit DNA replication and mitosis and promote DNA repair, recombination and apoptosis. Phosphorylates 'Ser-139' of histone variant H2AX/H2AFX at sites of DNA damage, thereby regulating DNA damage response mechanism. Required for FANCD2 ubiquitination. Critical for maintenance of fragile site stability and efficient regulation of centrosome duplication.

Tissue specificity

Ubiquitous, with highest expression in testis. Isoform 2 is found in pancreas, placenta and liver but not in heart, testis and ovary.

Involvement in disease

Defects in ATR are a cause of Seckel syndrome type 1 (SCKL1) [MIM:210600]. SCKL1 is a rare autosomal recessive disorder characterized by growth retardation, microcephaly with mental retardation, and a characteristic 'bird-headed' facial appearance.

Sequence similarities

Belongs to the PI3/P14-kinase family. ATM subfamily.
Contains 1 FAT domain.
Contains 1 FATC domain.
Contains 2 HEAT repeats.
Contains 1 PI3K/PI4K domain.

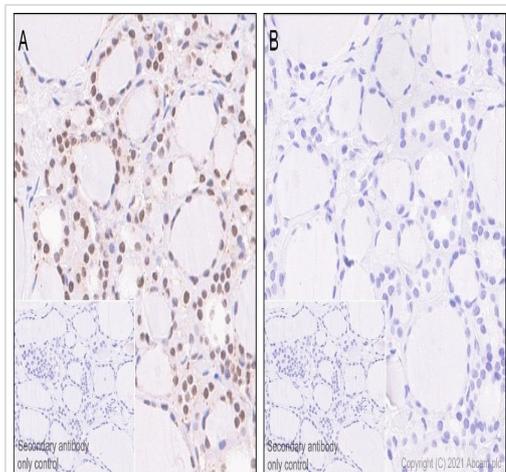
Post-translational modifications

Phosphorylated; autophosphorylates in vitro.

Cellular localization

Nucleus. Nucleus > PML body. Depending on the cell type, it can also be found in PML nuclear bodies. Recruited to chromatin during S-phase. Redistributes to discrete nuclear foci upon DNA damage, hypoxia or replication fork stalling.

Images

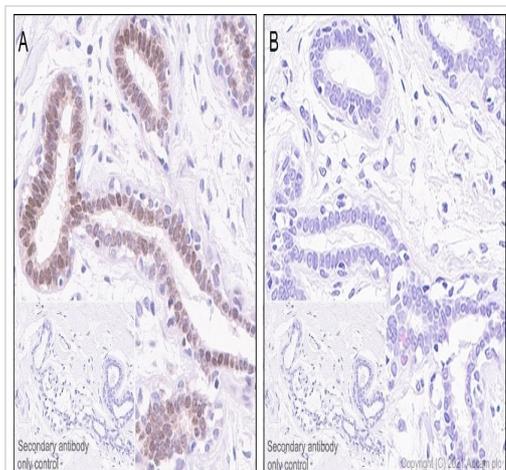


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATR (phospho S428) antibody [EPR2184] (ab178407)

Immunohistochemical analysis of Paraffin-embedded human thyroid tissue sections labelling ATR with ab178407 at 1/5000 dilution followed by ready to use secondary ab209101 Rabbit specific IHC polymer detection kit HRP/DAB. Counterstained with Hematoxylin. Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 20 minutes.

Nuclear staining on human thyroid without alkaline phosphatase (or Lambda Protein Phosphatase) treatment; No signal was detected when tissues were treated with alkaline phosphatase (image B).

The section was incubated with ab178407 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument

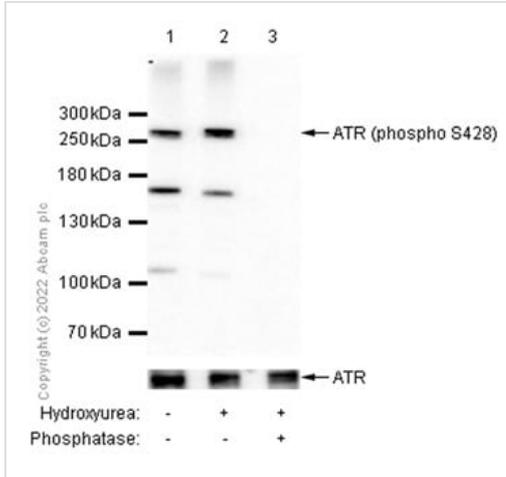


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATR (phospho S428) antibody [EPR2184] (ab178407)

Immunohistochemical analysis of Paraffin-embedded human breast tissue sections labelling ATR with ab178407 at 1/5000 dilution followed by ready to use secondary ab209101 Rabbit specific IHC polymer detection kit HRP/DAB. Counterstained with Hematoxylin. Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 20 minutes.

Nuclear staining on human breast without alkaline phosphatase (or Lambda Protein Phosphatase) treatment; No signal was detected when tissues were treated with alkaline phosphatase (image B).

The section was incubated with ab178407 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument



Western blot - Anti-ATR (phospho S428) antibody [EPR2184] (ab178407)

All lanes : Anti-ATR (phospho S428) antibody [EPR2184] (ab178407) at 1/1000 dilution

Lane 1 : Untreated HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 2 : HeLa treated with 4mM hydroxyurea for 20 hours whole cell lysate

Lane 3 : HeLa treated with 4mM hydroxyurea for 20 hours whole cell lysate, then the membrane treated with Alkaline Phosphatase for 1 hour

Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

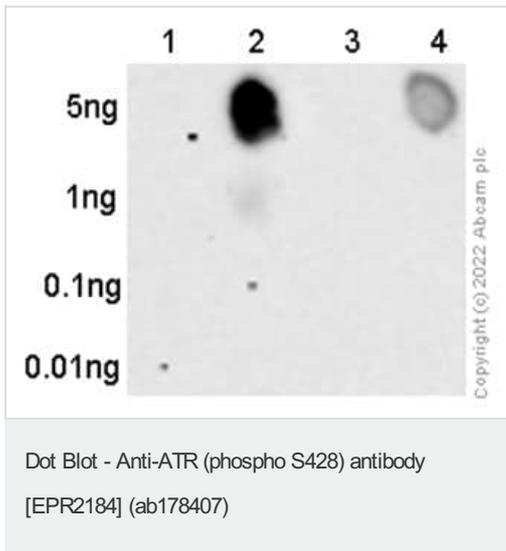
Predicted band size: 300 kDa

Observed band size: 270 kDa

Blocking and diluting buffer: 5% NFDM/TBST

Exposure time: 60 seconds

We are unsure of the nature of the 160kDa band.



Dot blot analysis using 1/1000 dilution ab178407 and Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) secondary at 1/100000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST

Lane 1: ATR non-phospho peptide

Lane 2: ATR S428 phospho peptide

Lane 3: ATR S435 phospho peptide

Lane 4: ATR S428+S435 phospho peptide

Exposure time: 3 minutes

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-ATR (phospho S428) antibody [EPR2184] (ab178407)

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

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