# abcam

# Product datasheet

# Anti-p95/NBS1 (phospho S343) antibody [EP178] ab109453



# 3 References 4 Images

#### Overview

Product name Anti-p95/NBS1 (phospho S343) antibody [EP178]

**Description** Rabbit monoclonal [EP178] to p95/NBS1 (phospho S343)

Host species Rabbit

Tested applications Suitable for: ICC/IF, Dot blot, WB

Unsuitable for: Flow Cyt or IHC-P

Species reactivity Reacts with: Human

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Jurkat, HeLa cell lysates (treated and untreated with Etopside). ICC/IF: Jurkat cells treated

with Etoposide (25uM) for 2 h

**General notes**This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

Improved sensitivity and specificityLong-term security of supply

- Animal-free production

For more information see here.

Our RabMAb $^{@}$  technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to  ${\hbox{\bf RabMAb}^{@}}$   ${\hbox{\bf patents}}$ .

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

#### **Properties**

Form Liquid

Storage instructions Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

Purity Protein A purified

**Clonality** Monoclonal

Clone number EP178

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee

Our Abpromise guarantee covers the use of ab109453 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
ICC/IF		1/50 - 1/100.
Dot blot		Use at an assay dependent concentration.
WB		1/500 - 1/1000. Detects a band of approximately 95 kDa (predicted molecular weight: 84 kDa).

**Application notes** 

Is unsuitable for Flow Cyt or IHC-P.

#### **Target**

#### **Function**

Component of the MRE11-RAD50-NBN (MRN complex) which plays a critical role in the cellular response to DNA damage and the maintenance of chromosome integrity. The complex is involved in double-strand break (DSB) repair, DNA recombination, maintenance of telomere integrity, cell cycle checkpoint control and meiosis. The complex possesses single-strand endonuclease activity and double-strand-specific 3'-5' exonuclease activity, which are provided by MRE11A. RAD50 may be required to bind DNA ends and hold them in close proximity. NBN modulate the DNA damage signal sensing by recruiting Pl3/Pl4-kinase family members ATM, ATR, and probably DNA-PKcs to the DNA damage sites and activating their functions. It can also recruit MRE11 and RAD50 to the proximity of DSBs by an interaction with the histone H2AX. NBN also functions in telomere length maintenance by generating the 3' overhang which serves as a primer for telomerase dependent telomere elongation. NBN is a major player in the control of intra-S-phase checkpoint and there is some evidence that NBN is involved in G1 and G2 checkpoints. The roles of NBS1/MRN encompass DNA damage sensor, signal transducer, and effector, which enable cells to maintain DNA integrity and genomic stability. Forms a complex with RBBP8 to link DNA double-strand break sensing to resection. Enhances AKT1 phosphorylation possibly by association with the mTORC2 complex.

Tissue specificity

Ubiquitous. Expressed at high levels in testis.

Involvement in disease

Nijmegen breakage syndrome

Breast cancer Aplastic anemia

Defects in NBN might play a role in the pathogenesis of childhood acute lymphoblastic leukemia

(ALL).

Sequence similarities

Contains 1 BRCT domain.

Contains 1 FHA domain.

**Domain** 

The FHA and BRCT domains are likely to have a crucial role for both binding to histone H2AFX

and for relocalization of MRE11/RAD50 complex to the vicinity of DNA damage.

The C-terminal domain contains a MRE11-binding site, and this interaction is required for the

nuclear localization of the MRN complex.

The EEXXXDDL motif at the C-terminus is required for the interaction with ATM and its recruitment to sites of DNA damage and promote the phosphorylation of ATM substrates, leading to the events of DNA damage response.

Post-translational modifications

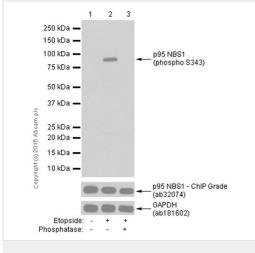
Phosphorylated by ATM in response of ionizing radiation, and such phosphorylation is responsible intra-S phase checkpoint control and telomere maintenance.

**Cellular localization** 

Nucleus. Nucleus, PML body. Chromosome, telomere. Localizes to discrete nuclear foci after

treatment with genotoxic agents.

# **Images**



Western blot - Anti-p95/NBS1 (phospho S343) antibody [EP178] (ab109453)

All lanes: Anti-p95/NBS1 (phospho S343) antibody [EP178] (ab109453) at 1/5000 dilution

Lane 1: Untreated HeLa (Human epithelial cell line from cervix adenocarcinoma) cells whole cell lysates

Lane 2: HeLa (Human epithelial cell line from cervix adenocarcinoma) cells were treated with Etopside whole cell lysates

Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) cells were treated with Etopside whole cell lysates. Then the membrane was incubated with Alkaline phosphatase.

Lysates/proteins at 15 µg per lane.

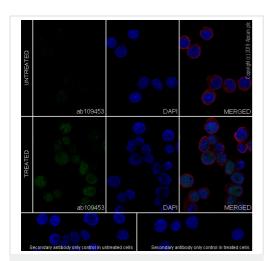
### Secondary

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

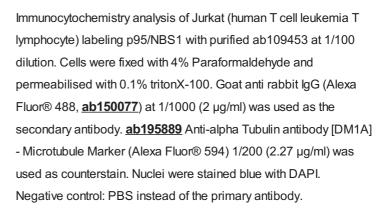
Predicted band size: 84 kDa Observed band size: 95 kDa

Exposure time: 1 minute

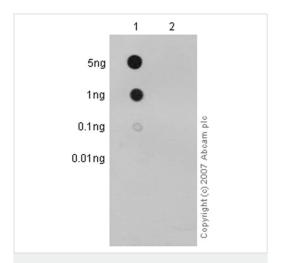
Blocking and diluting buffer: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Antip95/NBS1 (phospho S343) antibody [EP178] (ab109453)



Confocal image showing increased nuclear staining in Jurkat cells treated with Etoposide (25uM) for 2 h.

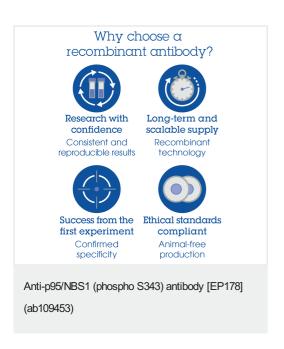


Dot Blot - Anti-p95/NBS1 (phospho S343) antibody [EP178] (ab109453)

Dot blot analysis of p95/NBS1 (pS343) peptide (Lane 1) and p95/NBS1 non-phospho peptide (Lane 2) labelling p95/NBS1 (phospho S343) with ab109453 at a dilution of 1/1000. A Peroxidase-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody at a dilution of 1/2500.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.



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