

# p-Nibrin (Ser 343)-R: sc-12936-R

## BACKGROUND

Nijmegen breakage syndrome (NBS) is characterized by extreme radiation sensitivity, chromosomal instability and cancer. These phenotypes are similar to those of ataxia telangiectasia mutated (ATM) disease, where there is a deficiency in a protein kinase that is activated by DNA damage, indicating that the NBS1 (Nibrin) and ATM proteins may participate in common pathways. Nibrin is specifically phosphorylated in response to  $\gamma$ -radiation, ultraviolet light and exposure to hydroxyurea. The phosphorylation of Nibrin requires catalytically active ATM. ATM physically interacts with and phosphorylates Nibrin on Serine 343 both *in vitro* and *in vivo*. Serine 343 is phosphorylated *in vitro* by ATM and the modification of this residue *in vivo* is essential for the cellular response to DNA damage. This response includes S-phase checkpoint activation, formation of the NBS1/Mre11/Rad50 nuclear foci and rescue of hypersensitivity to ionizing radiation.

## REFERENCES

- Kim, S.T., et al. 1999. Substrate specificities and identification of putative substrates of ATM kinase family members. *J. Biol. Chem.* 274: 37538-37543.
- Lim, D.S., et al. 2000. ATM phosphorylates p95/NBS1 in an S-phase checkpoint pathway. *Nature* 404: 613-617.
- Zhao, S., et al. 2000. Functional link between ataxia-telangiectasia and Nijmegen breakage syndrome gene products. *Nature* 405: 473-477.
- Wu, X., et al. 2000. ATM phosphorylation of Nijmegen breakage syndrome protein is required in a DNA damage response. *Nature* 405: 477-482.
- Gatei, M., et al. 2000. ATM-dependent phosphorylation of Nibrin in response to radiation exposure. *Nat. Genet.* 25: 115-119.

## CHROMOSOMAL LOCATION

Genetic locus: NBN (human) mapping to 8q21; Nbn (mouse) mapping to 4 A.

## SOURCE

p-Nibrin (Ser 343)-R is a rabbit polyclonal antibody raised against a short amino acid sequence containing phosphorylated Ser 343 of Nibrin of human origin.

## PRODUCT

Each vial contains 200  $\mu$ g IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-12936 P, (100  $\mu$ g peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

## STORAGE

Store at 4° C, \*\*DO NOT FREEZE\*\*. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

## RESEARCH USE

For research use only, not for use in diagnostic procedures.

## APPLICATIONS

p-Nibrin (Ser 343)-R is recommended for detection of Ser 343 phosphorylated Nibrin of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2  $\mu$ g per 100-500  $\mu$ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for Nibrin siRNA (h): sc-36061, Nibrin siRNA (m): sc-36062, Nibrin shRNA Plasmid (h): sc-36061-SH, Nibrin shRNA Plasmid (m): sc-36062-SH, Nibrin shRNA (h) Lentiviral Particles: sc-36061-V and Nibrin shRNA (m) Lentiviral Particles: sc-36062-V.

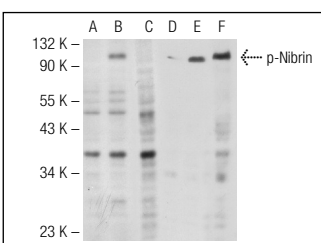
Molecular Weight of p-Nibrin: 95 kDa.

Positive Controls: HeLa whole cell lysate + ATM, Jurkat whole cell lysate: sc-2204 or HeLa whole cell lysate: sc-2200.

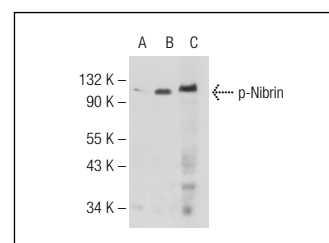
## RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Western Blotting Luminol Reagent: sc-2048 and Lambda Phosphatase: sc-200312A. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## DATA



Western blot analysis of Nibrin phosphorylation in untreated (A, D), UV irradiated (B, E) and UV irradiated and lambda protein phosphatase (sc-200312A) treated (C, F) HeLa whole cell lysates. Antibodies tested include p-Nibrin (Ser 343)-R: sc-12936-R (A, B, C) and Nibrin (107): sc-56166 (D, E, F).



p-Nibrin (Ser 343)-R: sc-12936-R. Western blot analysis of Nibrin phosphorylation in untreated (A), UV irradiated (B) and UV irradiated and lambda protein phosphatase (sc-200312A) treated (C) HeLa whole cell lysates.

## SELECT PRODUCT CITATIONS

- Austen, B., et al. 2005. Mutations in the ATM gene lead to impaired overall and treatment-free survival that is independent of IGVH mutation status in patients with B-CLL. *Blood* 106: 3175-3182.
- Rink, L., et al. 2007. Enhanced phosphorylation of NBS1, a member of DNA repair/checkpoint complex Mre11-Rad50-NBS1, can be targeted to increase the efficacy of imatinib mesylate against BCR/ABL-positive leukemia cells. *Blood* 110: 651-660.