

# p-nestin (A-4): sc-377538

## BACKGROUND

Nestin is a major intermediate filament (IF) protein of embryonic central nervous system progenitor cells. Nestin is also a component of the dynamic IF network during muscle development where it polymerizes with desmin and vimentin. Nestin co-assembles with vimentin or  $\alpha$ -internexin and forms heterodimer coiled-coil molecules which then further assemble into 10 nm IFs. Deletion of the IF consensus rod domain in nestin alters nestin localization in CNS precursor cells and radial glial cells *in vivo*. Nestin is a marker for neuroepithelial stem cells, glioma cells, and tumor endothelial cells during rapid growth. During axon elongation of differentiation neurons, nestin localizes to the growth cones and may play a role in growth cone guidance. In the rat adrenal gland, nestin is expressed by the zona fasciculata and the zona reticularis. Nestin is also expressed by dermatomal cells and by myoblasts during the earliest stages of myogenesis. Thr 316 is a cdc2-specific phosphorylation site in Nestin. Phosphorylation at this site is increased during mitosis and is used as a marker for Cdk5-specific phosphorylation. Cdk5 and cdc2 induces phosphorylation at both residues Thr 316 and Thr 1583.

## CHROMOSOMAL LOCATION

Genetic locus: NES (human) mapping to 1q23.1; Nes (mouse) mapping to 3 F1.

## SOURCE

p-nestin (A-4) is a mouse monoclonal antibody epitope corresponding to a short amino acid sequence containing Thr 316 phosphorylated nestin of rat origin.

## PRODUCT

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

Blocking peptide available for competition studies, sc-377538 P, (100  $\mu$ g peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

## APPLICATIONS

p-nestin (A-4) is recommended for detection of Thr 316 phosphorylated nestin of mouse, rat and human origin by Western Blotting (starting dilution 1:100, 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).

p-nestin (A-4) is also recommended for detection of correspondingly phosphorylated Nestin in additional species, including bovine and porcine.

Suitable for use as control antibody for nestin siRNA (h): sc-36032, nestin siRNA (m): sc-36033, nestin shRNA Plasmid (h): sc-36032-SH, nestin shRNA Plasmid (m): sc-36033-SH, nestin shRNA (h) Lentiviral Particles: sc-36032-V and nestin shRNA (m) Lentiviral Particles: sc-36033-V.

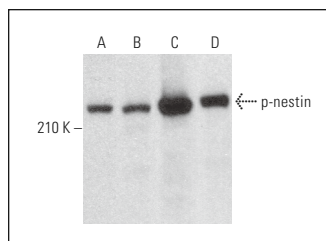
Molecular Weight of p-nestin: 190-200 kDa.

Positive Controls: SK-N-SH cell lysate: sc-2410, U-251-MG whole cell lysate: sc-364176 or F9 cell lysate: sc-2245.

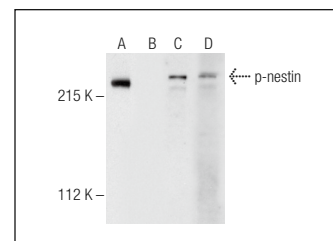
## RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Lambda Phosphatase: sc-200312A and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein L-Agarose: sc-2336 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

## DATA



p-nestin (A-4): sc-377538. Western blot analysis of p-nestin expression in U-251-MG (A), SK-N-SH (B), IMR-32 (C) and F9 (D) whole cell lysates.



Western blot analysis of nestin phosphorylation in untreated (A, C) and lambda protein phosphatase (sc-200312A) treated (B, D) SH-SY5Y whole cell lysates. Antibodies tested include p-nestin (A-4): sc-377538 (A, B) and nestin (C-20): sc-21247 (C, D).

## SELECT PRODUCT CITATIONS

- Wang, Y., et al. 2023. The intermediate filament protein nestin serves as a molecular hub for smooth muscle cytoskeletal signaling. *Respir. Res.* 24: 157.

## 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.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.



See **nestin (Rat-401): sc-33677** for nestin antibody conjugates, including AC, HRP, FITC, PE, and Alexa Fluor® 488, 546, 594, 647, 680 and 790.