p-NF-H (RNF404): sc-32728



The Power to Question

BACKGROUND

Neurofilament-H (for neurofilament heavy polypeptide, or NF-H), a member of the intermediate filament family, is a major component of neuronal cytoskeletons. Neurofilaments are dynamic structures; they contain phosphorylation sites for a large number of protein kinases, including protein kinase A, protein kinase C, cyclin-dependent kinase 5, extracellular signal regulated kinase, glycogen synthase kinase-3, and stress-activated protein kinase γ . In addition to their role in the control of axon caliber, neurofilaments may affect other cytoskeletal elements, such as microtubules and actin filaments. Changes in neurofilament phosphorylation or metabolism are frequently observed in neurodegenerative diseases, including amotrophic lateral sclerosis (ALS), Parkinson's disease, and Alzheimer's disease.

REFERENCES

- Mattei, M.G., et al. 1988. The gene encoding the large human neurofilament subunit (NF-H) maps to the q121-q131 region on human chromosome 22. Hum. Genet. 80: 293-295.
- Angelides, K.J., et al. 1989. Assembly and exchange of intermediate filament proteins of neurons: neurofilaments are dynamic structures. J. Cell Biol. 108: 1495-1506.
- Sihag, R.K., et al. 1989. *In vivo* phosphorylation of distinct domains of the 70 kilodalton neurofilament subunit involves different protein kinases. J. Biol. Chem. 264: 457-464.
- 4. Hisanaga, S., et al. 1990. Effects of phosphorylation of the neurofilament L protein on filamentous structures. Cell Regul. 1: 237-248.
- Gonda, Y., et al. 1990. Involvement of protein kinase C in the regulation of assembly-disassembly of neurofilaments in vitro. Biochem. Biophys. Res. Commun. 167: 1316-1325.
- Lukas, Z., et al. 1993. Expression of phosphorylated high molecular weight neurofilament protein (NF-H) and Vimentin in human developing dorsal root ganglia and spinal cord. Histochemistry 100: 495-502.
- 7. Nakamura, Y, et al. 1997. Abnormal distribution of neurofilament L in neurons with Alzheimer's disease. Neurosci. Lett. 225: 201-204.
- 8. Nakamura, Y., et al. 1999. Casein kinase II is responsible for phosphorylation of NF-L at Ser-473. FEBS Lett. 455: 83-86.
- 9. Cunningham, C., et al. 2005. Central and systemic endotoxin challenges exacerbate the local inflammatory response and increase neuronal death during chronic neurodegeneration. J. Neurosci. 25: 9275-9284.

CHROMOSOMAL LOCATION

Genetic locus: NEFH (human) mapping to 22q12.2; Nefh (mouse) mapping to 11 A1.

SOURCE

p-NF-H (RNF404) is a mouse monoclonal antibody raised against a neurofilament NF-H protein isolated from a cytoskeletal preparation from brain tissue homogenate of calf origin.

PRODUCT

Each vial contains 200 μg IgG_{2a} kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

APPLICATIONS

p-NF-H (RNF404) is recommended for detection of phosphorylated NF-H of mouse, rat, human, *Xenopus laevis* and bovine origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)] and immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

Suitable for use as control antibody for NF-H siRNA (h): sc-42068, NF-H siRNA (m): sc-42069, NF-H shRNA Plasmid (h): sc-42068-SH, NF-H shRNA Plasmid (m): sc-42069-SH, NF-H shRNA (h) Lentiviral Particles: sc-42068-V and NF-H shRNA (m) Lentiviral Particles: sc-42069-V.

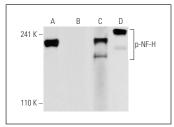
Molecular Weight of p-NF-H: 200 kDa.

Positive Controls: NF-H (h): 293T Lysate: sc-111457, rat brain extract: sc-2392 or mouse brain extract: sc-2253.

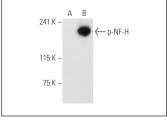
RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM 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 A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG κ 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



Western blot analysis of NF-H phosphorylation in untreated (**A,C**) and lambda protein phosphatase (sc-200312A) treated (**B,D**) rat brain tissue extracts. Antibodies tested include p-NF-H (RNF404): sc-32728 (**A,B**) and NF-H (2D2): sc-20014 (**C,D**).



p-NF-H (RNF404): sc-32728. Western blot analysis of NF-H prosphorylation in non-transfected: sc-117752 (**A**) and human NF-H transfected: sc-111457 (**B**) 293T whole cell I vsates.

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.