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Neurabin-II shRNA (h) Lentiviral Particles: sc-43962-V

BACKGROUND

Neurabin-II, also called spinophilin, interacts with actin and PP-1 in dendritic spines of the central nervous system. The gene encoding human neurabin-II maps to chromosome 17q21.33. The structural characteristics of neurabin-II include one F-actin binding domain at the N-terminal region, a predicted coiled-coil structure at the C-terminal, one PDZ domain at the middle region, and a domain known to interact with transmembrane proteins. Neurabin-II bundles actin filaments *in vitro*. *In vivo*, spinophilin localizes to the cortical sites of actin filaments and to the sites of active membrane remodeling. Neurabin-II also forms a complex with the catalytic subunit of PP1 and modulates PP1 enzymatic activity *in vitro*. Neurabin-II localizes to the head of dendritic spines and aids in the ability of PP-1 to regulate the activity of α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) and N-methyl-D-aspartate (NMDA) receptors. In this manner, neurabin-II modulates both glutamatergic synaptic transmission and dendritic morphology. Synergistic interactions between spinophilin and human tumor suppressor ARF suggest a role for neurabin-II in cell growth.

REFERENCES

- Allen, P.B., et al. 1997. Spinophilin, a novel protein phosphatase 1 binding protein localized to dendritic spines. *Proc. Natl. Acad. Sci. USA* 94: 9956-9961.
- Satoh, A., et al. 1998. Neurabin-II/spinophilin. An actin filament-binding protein with one pdz domain localized at cadherin-based cell-cell adhesion sites. *J. Biol. Chem.* 273: 3470-3475.
- Feng, J., et al. 2000. Spinophilin regulates the formation and function of dendritic spines. *Proc. Natl. Acad. Sci. USA* 97: 9287-9292.
- Stephens, D.J., et al. 2000. *In vivo* dynamics of the F-actin-binding protein neurabin-II. *Biochem. J.* 345: 185-194.

CHROMOSOMAL LOCATION

Genetic locus: PPP1R9B (human) mapping to 17q21.33.

PRODUCT

Neurabin-II shRNA (h) Lentiviral Particles are concentrated, transduction-ready viral particles containing a target-specific construct that encodes a 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Neurabin-II siRNA (h): sc-43962 and Neurabin-II shRNA Plasmid (h): sc-43962-SH as alternate gene silencing products.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

APPLICATIONS

Neurabin-II shRNA (h) Lentiviral Particles is recommended for the inhibition of Neurabin-II expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Neurabin-II (D-7): sc-373974 is recommended as a control antibody for monitoring of Neurabin-II gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Neurabin-II gene expression knockdown using RT-PCR Primer: Neurabin-II (h)-PR: sc-43962-PR (20 μ l, 479 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.