

Neurofibromin siRNA (r): sc-270621

BACKGROUND

Neurofibromatosis type 1 (NF1), or von Recklinghausen neurofibromatosis, is one of the most common autosomal dominant disorders in humans. Early linkage analysis mapped the NF1 gene to chromosome 17. The predicted NF1 transcript encodes the 2,818 amino acid protein Neurofibromin, also designated NF1-GAP-related protein (NF1GRP). By sequence analysis, similarity has been demonstrated within a small region of Neurofibromin and members of the Ras GAP gene family. Functionally, Neurofibromin has been shown by biochemical analysis involving Ras-GAP hydrolysis and functional complementation in yeast to further resemble GAP protein. The Neurofibromin protein is expressed at relatively constant levels in a broad range of cell lines and tissues including brain, lung, liver, kidney, spleen, muscle and colon. Although little is known regarding the function of Neurofibromin, the homology with the catalytic domain of proteins with GTPase activity suggests that Neurofibromin may also interact *in vivo* with Ras proteins.

REFERENCES

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4. Martin, G.A., et al. 1990. The GAP-related domain of the neurofibromatosis type 1 gene product interacts with Ras p21. *Cell* 63: 843-849.
5. Xu, G., et al. 1990. The catalytic domain of the neurofibromatosis type 1 gene product stimulates ras GTPase and complements *ira* mutants of *S. cerevisiae*. *Cell* 63: 835-841.
6. Gutmann, D.H., et al. 1991. Identification of the neurofibromatosis type 1 gene product. *Proc. Natl. Acad. Sci. USA* 88: 9658-9662.
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CHROMOSOMAL LOCATION

Genetic locus: Nf1 (rat) mapping to 10q25.

PRODUCT

Neurofibromin siRNA (r) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Neurofibromin shRNA Plasmid (r): sc-270621-SH and Neurofibromin shRNA (r) Lentiviral Particles: sc-270621-V as alternate gene silencing products.

For independent verification of Neurofibromin (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270621A, sc-270621B and sc-270621C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Neurofibromin siRNA (r) is recommended for the inhibition of Neurofibromin expression in rat cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

Neurofibromin (H-12): sc-376886 is recommended as a control antibody for monitoring of Neurofibromin 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 reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Neurofibromin gene expression knockdown using RT-PCR Primer: Neurofibromin (r)-PR: sc-270621-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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

PROTOCOLS

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