

Nup50 siRNA (h): sc-75979

BACKGROUND

Nuclear pore complexes (NPCs) are the channels for the bi-directional movement of macromolecules between the nucleus and cytoplasm, and contain more than 100 different subunits. Many of them belong to a family called nucleoporins, which are characterized by the presence of O-linked N-acetylglucosamine moieties and a distinctive pentapeptide repeat (XFXFG). Nup50 (nucleoporin 50), also known as NPAP60 or NPAP60L (nuclear pore-associated protein 60 kDa-like), is a 468 amino acid nuclear protein that functions as a binding site for export receptor-cargo complexes. Localizing to the nucleoplasmic fibrils of the nuclear pore complex, Nup50 associates with various transport receptor proteins including p27. While ubiquitously expressed, Nup50 is found at highest levels in peripheral blood leukocytes, testis and fetal liver, and contains multiple FG repeats in addition to a single RanBD1 domain.

REFERENCES

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3. Smitherman, M., et al. 2000. Characterization and targeted disruption of murine Nup50, a p27^{Kip1}-interacting component of the nuclear pore complex. *Mol. Cell. Biol.* 20: 5631-5642.
4. Lindsay, M.E., et al. 2002. Npap60/Nup50 is a tri-stable switch that stimulates importin- α : β -mediated nuclear protein import. *Cell* 110: 349-360.
5. Swaminathan, S. and Melchior, F. 2002. Nucleocytoplasmic transport: more than the usual suspects. *Dev. Cell* 3: 304-306.
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CHROMOSOMAL LOCATION

Genetic locus: NUP50 (human) mapping to 22q13.31.

PRODUCT

Nup50 siRNA (h) 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 Nup50 shRNA Plasmid (h): sc-75979-SH and Nup50 shRNA (h) Lentiviral Particles: sc-75979-V as alternate gene silencing products.

For independent verification of Nup50 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-75979A, sc-75979B and sc-75979C.

RESEARCH USE

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

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

Nup50 siRNA (h) is recommended for the inhibition of Nup50 expression in human 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

Nup50 (G-4): sc-398993 is recommended as a control antibody for monitoring of Nup50 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 λ BP-HRP: sc-516132 or m-IgG λ BP-HRP (Cruz Marker): sc-516132-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 λ BP-FITC: sc-516185 or m-IgG λ BP-PE: sc-516186 (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 Nup50 gene expression knockdown using RT-PCR Primer: Nup50 (h)-PR: sc-75979-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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