

NHPX siRNA (m): sc-149964

BACKGROUND

NHPX, also known as NHP2L1 (NHP2 non-histone chromosome protein 2-like 1), FA-1, 15.5K, OTK27, SNU13, SPAG12 or SNRNP15-5, is a 128 amino acid protein belonging to the ribosomal protein L7Ae family. NHPX localizes to the nucleus, mainly concentrated in the dense fibrillar component of the nucleolus. Ubiquitously expressed, NHPX binds to the 5'-stem-loop of U4 snRNA and may be involved in the late stage of spliceosome assembly. Following RNA binding, NHPX undergoes a conformational change and is recruited to introns, where NHPX is required for the subsequent recruitment of PRPF31 and the activation of the spliceosome complex. NHPX is expressed as two isoforms produced by alternative splicing.

REFERENCES

1. Vidovic, I., Nottrott, S., Hartmuth, K., Lührmann, R. and Ficner, R. 2000. Crystal structure of the spliceosomal 15.5 kD protein bound to a U4 snRNA fragment. *Mol. Cell* 6: 1331-1342.
2. Leung, A.K. and Lamond, A.I. 2002. *In vivo* analysis of NHPX reveals a novel nucleolar localization pathway involving a transient accumulation in splicing speckles. *J. Cell Biol.* 157: 615-629.
3. Scherl, A., Coute, Y., Deon, C., Calle, A., Kindbeiter, K., Sanchez, J.C., Greco, A., Hochstrasser, D. and Diaz, J.J. 2002. Functional proteomic analysis of human nucleolus. *Mol. Biol. Cell* 13: 4100-4109.
4. Watkins, N.J., Dickmanns, A. and Lührmann, R. 2002. Conserved stem II of the box C/D motif is essential for nucleolar localization and is required, along with the 15.5K protein, for the hierarchical assembly of the box C/D snoRNP. *Mol. Cell. Biol.* 22: 8342-8352.
5. Naz, R.K. and Zhu, X. 2002. Molecular cloning and sequencing of cDNA encoding for human FA-1 antigen. *Mol. Reprod. Dev.* 63: 256-268.
6. Zhou, Z., Licklider, L.J., Gygi, S.P. and Reed, R. 2002. Comprehensive proteomic analysis of the human spliceosome. *Nature* 419: 182-185.
7. Andersen, J.S., Lam, Y.W., Leung, A.K., Ong, S.E., Lyon, C.E., Lamond, A.I. and Mann, M. 2005. Nucleolar proteome dynamics. *Nature* 433: 77-83.
8. Soss, S.E. and Flynn, P.F. 2007. Functional implications for a prototypical K-turn binding protein from structural and dynamical studies of 15.5K. *Biochemistry* 46: 14979-14986.
9. McKeegan, K.S., Debieux, C.M., Boulon, S., Bertrand, E. and Watkins, N.J. 2007. A dynamic scaffold of pre-snoRNP factors facilitates human box C/D snoRNP assembly. *Mol. Cell. Biol.* 27: 6782-6793.

CHROMOSOMAL LOCATION

Genetic locus: Nhp2l1 (mouse) mapping to 15 E1.

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.

PRODUCT

NHPX siRNA (m) is a pool of 2 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 NHPX shRNA Plasmid (m): sc-149964-SH and NHPX shRNA (m) Lentiviral Particles: sc-149964-V as alternate gene silencing products.

For independent verification of NHPX (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-149964A and sc-149964B.

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

NHPX siRNA (m) is recommended for the inhibition of NHPX expression in mouse 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NHPX gene expression knockdown using RT-PCR Primer: NHPX (m)-PR: sc-149964-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.