

p54/nrb siRNA (h): sc-38163

BACKGROUND

Found in both primary and transformed human cells, paraspeckles are discrete bodies in the interchromatin nucleoplasmic space which contain p54/nrb (nuclear RNA-binding protein) and at least two other RNA-binding proteins, paraspeckle protein 1 (PSP1) and paraspeckle protein 2 (PSP2). Paraspeckles often co-localize with splicing speckles, which are the site of splicing factor accumulation. Paraspeckle proteins, including p54/nrb, move dynamically between the nucleolus and paraspeckles and translocate to distinct caps in the nucleolar periphery when transcription is inhibited. Originally purified from HeLa cells, the nuclear p54/nrb has two RNA recognition motifs and shares extensive homology with both the human splicing factor PSF and *Drosophila* NONA/BJ6, which is required for normal vision and courtship. The shared domain between these proteins is termed a DBHS (*Drosophila* behavior, human splicing) domain and may play a role in regulating various pathways at the level of pre-mRNA splicing.

REFERENCES

1. Dong, B., et al. 1993. Purification and cDNA cloning of HeLa cell p54^{nrb}, a nuclear protein with two RNA recognition motifs and extensive homology to human splicing factor PSF and *Drosophila* NONA/BJ6. *Nucleic Acids Res.* 21: 4085-4092.
2. Brown, C.J., et al. 1997. Expression of genes from the human active and inactive X chromosomes. *Am. J. Hum. Genet.* 60: 1333-1343.
3. Zhang, Z. and Carmichael, G.G. 2001. The fate of dsRNA in the nucleus: a p54^{nrb}-containing complex mediates the nuclear retention of promiscuously A-to-I edited RNAs. *Cell* 106: 465-475.

CHROMOSOMAL LOCATION

Genetic locus: NONO (human) mapping to Xq13.1.

PRODUCT

p54/nrb 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 p54/nrb shRNA Plasmid (h): sc-38163-SH and p54/nrb shRNA (h) Lentiviral Particles: sc-38163-V as alternate gene silencing products.

For independent verification of p54/nrb (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-38163A, sc-38163B and sc-38163C.

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

p54/nrb siRNA (h) is recommended for the inhibition of p54/nrb 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

p54/nrb (F-5): sc-376804 is recommended as a control antibody for monitoring of p54/nrb 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 p54/nrb gene expression knockdown using RT-PCR Primer: p54/nrb (h)-PR: sc-38163-PR (20 μ l, 543 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ho, T.T., et al. 2016. Regulation of PCGEM1 by p54/nrb in prostate cancer. *Sci. Rep.* 6: 34529.
2. Rhee, D.K., et al. 2017. SFPQ, a multifunctional nuclear protein, regulates the transcription of PDE3A. *Biosci. Rep.* 37: BSR20170975.
3. Beeharay, Y., et al. 2018. The Hepatitis Delta Virus accumulation requires paraspeckle components and affects NEAT1 level and PSP1 localization. *Sci. Rep.* 8: 6031.

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.