SANTA CRUZ BIOTECHNOLOGY, INC.

PRPF3 siRNA (h): sc-88819

BACKGROUND

PRPF3 (PRP3 pre-mRNA processing factor 3 homolog), also known as RP18, PRP3, Prp3p, HPRP3 or HPRP3P, is an evolutionarily conserved protein involved in pre-mRNA splicing and functions as a component of the U4/U6.U5 tri-snRNP (small nuclear ribonucleoprotein) complex. Ubiquitously expressed with predominant expression in retina, blood, kidney and liver, PRPF3 localizes to nuclear speckles and is phosphorylated *in vitro*. PRPF3 directly interacts with PRPF4 and is present in the inactive spliceosome but is not found in the catalytically active spliceosome. Mutations in the gene encoding PRPF3 result in autosomal dominant retinitis pigmentosa type 18 (RP18), which leads to photoreceptor cell degeneration. RP18 patients initially exhibit a loss of their midperipheral visual field as well as night vision blindness. The disease eventually progresses to the loss of far peripheral visual field and finally the loss of central vision. This suggests that PRPF3 is a key player in the pre-mRNA splicing of photoreceptor-specific genes.

REFERENCES

1. Wang, A., et al. 1997. Identification and characterization of human genes encoding Hprp3p and Hprp4p, interacting components of the spliceosome. Hum. Mol. Genet. 6: 2117-2126.
2. Lauber, J., et al. 1997. The human U4/U6 snRNP contains 60 and 90kD proteins that are structurally homologous to the yeast splicing factors Prp4p and Prp3p. RNA 3: 926-941.
3. Horowitz, D.S., et al. 1997. A new cyclophilin and the human homologues of yeast Prp3 and Prp4 form a complex associated with U4/U6 snRNPs. RNA 3: 1374-1387.
4. Chakarova, C.F., et al. 2002. Mutations in HPRP3, a third member of pre-mRNA splicing factor genes, implicated in autosomal dominant retinitis pigmentosa. Hum. Mol. Genet. 11: 87-92.
5. Gonzalez-Santos, J.M., et al. 2002. Central region of the human splicing factor Hprp3p interacts with Hprp4p. J. Biol. Chem. 277: 23764-23772.
6. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 607301. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: PRPF3 (human) mapping to 1q21.2.

PRODUCT

PRPF3 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PRPF3 shRNA Plasmid (h): sc-88819-SH and PRPF3 shRNA (h) Lentiviral Particles: sc-88819-V as alternate gene silencing products.

For independent verification of PRPF3 (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-88819A, sc-88819B and sc-88819C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PRPF3 siRNA (h) is recommended for the inhibition of PRPF3 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 µM in 66 µl. 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

PRPF3 (B-1): sc-518271 is recommended as a control antibody for monitoring of PRPF3 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 PRPF3 gene expression knockdown using RT-PCR Primer: PRPF3 (h)-PR: sc-88819-PR (20 µl). 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.