

RP2 siRNA (h): sc-76428

BACKGROUND

RP2 (retinitis pigmentosa 2), also known as TBCCD2, is a 350 amino acid protein that localizes to the cytoplasmic side of the cell membrane and belongs to the TBCC family. Expressed ubiquitously, RP2 functions to stimulate the GTPase activity of tubulin and is thought to act as a guanine nucleotide dissociation inhibitor for ARL3 (ADP-ribosylation factor-like 3), preventing the GTP-bound form of ARL3 from dissociating. Via its ability to stimulate tubulin activity, RP2 plays an important role in retinal development. RP2 contains one C-CAP/cofactor C-like domain and can be myristoylated or palmitoylated, both of which are thought to be required for proper membrane targeting. Defects in the gene encoding RP2 are the cause of retinitis pigmentosa type 2 (RP2), a disorder characterized by the degeneration of photoreceptor cells, resulting in night vision blindness and an eventual loss of both peripheral and central vision.

REFERENCES

1. Thiselton, D.L., et al. 1996. Mapping the RP2 locus for X-linked retinitis pigmentosa on proximal Xp: a genetically defined 5-cM critical region and exclusion of candidate genes by physical mapping. *Genome Res.* 6: 1093-1102.
2. Schwahn, U., et al. 1998. Positional cloning of the gene for X-linked retinitis pigmentosa 2. *Nat. Genet.* 19: 327-332.
3. Rosenberg, T., et al. 1999. Genotype-phenotype correlation in X-linked retinitis pigmentosa 2 (RP2). *Ophthalmic Genet.* 20: 161-172.
4. Chapple, J.P., et al. 2000. Mutations in the N-terminus of the X-linked retinitis pigmentosa protein RP2 interfere with the normal targeting of the protein to the plasma membrane. *Hum. Mol. Genet.* 9: 1919-1926.
5. Thiselton, D.L., et al. 2000. Novel frameshift mutations in the RP2 gene and polymorphic variants. *Hum. Mutat.* 15: 580.
6. Breuer, D.K., et al. 2002. A comprehensive mutation analysis of RP2 and RPGR in a North American cohort of families with X-linked retinitis pigmentosa. *Am. J. Hum. Genet.* 70: 1545-1554.
7. Bartolini, F., et al. 2002. Functional overlap between retinitis pigmentosa 2 protein and the Tubulin-specific chaperone cofactor C. *J. Biol. Chem.* 277: 14629-14634.

CHROMOSOMAL LOCATION

Genetic locus: RP2 (human) mapping to Xp11.23.

PRODUCT

RP2 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 RP2 shRNA Plasmid (h): sc-76428-SH and RP2 shRNA (h) Lentiviral Particles: sc-76428-V as alternate gene silencing products.

For independent verification of RP2 (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-76428A, sc-76428B and sc-76428C.

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

RP2 siRNA (h) is recommended for the inhibition of RP2 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

RP2 (C-9): sc-390220 is recommended as a control antibody for monitoring of RP2 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 RP2 gene expression knockdown using RT-PCR Primer: RP2 (h)-PR: sc-76428-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.