

RD3 siRNA (h): sc-88397

BACKGROUND

Leber congenital amaurosis (LCA) is one of the most common causes of hereditary blindness or severe visual impairment in infants. Mutations in several genes with diverse functions mapping to two loci have been implicated in LCA causation. These proteins are involved in processes such as photoreceptor development and maintenance, phototransduction, vitamin A metabolism and protein trafficking. RD3 (retinal degeneration 3), also known as LCA12, is a 195 amino acid protein expressed in retina. RD3 is suggested to be part of the subnuclear protein complexes involved in diverse processes, such as transcription and splicing. Defects in the gene encoding RD3 are the cause of Leber congenital amaurosis type 12. Infants affected with Leber congenital amaurosis type 12 have little or no retinal photoreceptor function.

REFERENCES

1. Chang, B., et al. 2002. Retinal degeneration mutants in the mouse. *Vision Res.* 42: 517-525.
2. Mohamed, M.D., et al. 2003. Progression of phenotype in Leber's congenital amaurosis with a mutation at the LCA5 locus. *Br. J. Ophthalmol.* 87: 473-475.
3. Friedman, J.S., et al. 2006. Premature truncation of a novel protein, RD3, exhibiting subnuclear localization is associated with retinal degeneration. *Am. J. Hum. Genet.* 79: 1059-1070.
4. Gerber, S., et al. 2007. Mutations in LCA5 are an uncommon cause of Leber congenital amaurosis (LCA) type II. *Hum. Mutat.* 28: 1245.
5. Ramprasad, V.L., et al. 2008. Identification of a novel splice-site mutation in the Lebercilin (LCA5) gene causing Leber congenital amaurosis. *Mol. Vis.* 14: 481-486.
6. den Hollander, A.I., et al. 2008. Leber congenital amaurosis: genes, proteins and disease mechanisms. *Prog. Retin. Eye Res.* 27: 391-419.
7. Kukekova, A.V., et al. 2009. Canine RD3 mutation establishes rod-cone dysplasia type 2 (rcd2) as ortholog of human and murine rd3. *Mamm. Genome* 20: 109-123.

CHROMOSOMAL LOCATION

Genetic locus: RD3 (human) mapping to 1q32.3.

PRODUCT

RD3 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 RD3 shRNA Plasmid (h): sc-88397-SH and RD3 shRNA (h) Lentiviral Particles: sc-88397-V as alternate gene silencing products.

For independent verification of RD3 (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-88397A, sc-88397B and sc-88397C.

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

RD3 siRNA (h) is recommended for the inhibition of RD3 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

RD3 (A-9): sc-390653 is recommended as a control antibody for monitoring of RD3 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 RD3 gene expression knockdown using RT-PCR Primer: RD3 (h)-PR: sc-88397-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.