

ABCA4 siRNA (h): sc-41140

BACKGROUND

The ATP-binding cassette (ABC) superfamily is comprised of transmembrane proteins involved in energy-dependent transport of a variety of substrates across membranes. ABCA4 (also designated ABCR, photoreceptor RIM or RMP) is a photoreceptor specific ATP-binding cassette (ABC) transporter. ABCA4 is exclusively expressed within photoreceptor cells, indicating that ABCA4 mediates the transport of an essential molecule either into or out of photoreceptor cells. Mutations in the gene encoding ABCA4 are responsible for autosomal recessive Stargardt disease (STGD), an early onset macular degeneration, and some forms of autosomal recessive cone-rod dystrophy and autosomal recessive retinitis pigmentosa. In addition, heterozygosity for ABCA4 mutations may possess a risk factor for age-related macular degeneration. ABCA4 is most closely related to the mouse and human ABC1 and ABC2 and maps to human chromosome 1p22.1.

REFERENCES

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- Dean, M. and Allikmets, R. 1995. Evolution of ATP-binding cassette transporter genes. *Curr. Opin. Genet. Dev.* 5: 779-785.
- Allikmets, R., et al. 1997. Mutation of the Stargardt disease gene (ABCR) in age-related macular degeneration. *Science* 277: 1805-1807.
- Schwiebert, E.M. 1999. ABC transporter-facilitated ATP conductive transport. *Am. J. Physiol.* 276: 1-8.
- Sun, H., et al. 2000. Biochemical defects in ABCR protein variants associated with human retinopathies. *Nat. Genet.* 26: 242-246.
- Yatsenko, A.N., et al. 2001. Late-onset Stargardt disease is associated with missense mutations that map outside known functional regions of ABCR (ABCA4). *Hum. Genet.* 108: 346-355.
- Paloma, E., et al. 2001. Spectrum of ABCA (ABCR) gene mutations in Spanish patients with autosomal recessive macular dystrophies. *Hum. Mutat.* 17: 504-510.

CHROMOSOMAL LOCATION

Genetic locus: ABCA4 (human) mapping to 1p22.1.

PRODUCT

ABCA4 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 ABCA4 shRNA Plasmid (h): sc-41140-SH and ABCA4 shRNA (h) Lentiviral Particles: sc-41140-V as alternate gene silencing products.

For independent verification of ABCA4 (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-41140A, sc-41140B and sc-41140C.

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

ABCA4 siRNA (h) is recommended for the inhibition of ABCA4 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

ABCA4 (3F4): sc-65672 is recommended as a control antibody for monitoring of ABCA4 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 ABCA4 gene expression knockdown using RT-PCR Primer: ABCA4 (h)-PR: sc-41140-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.