

βB2-crystallin siRNA (h): sc-40444

BACKGROUND

Crystallins are the major proteins of the vertebrate eye lens, where they maintain the transparency and refractive index of the lens. Crystallins are divided into α , β and γ families, and the β - and γ -crystallins also comprise a superfamily. Crystallins usually contain seven distinctive protein regions, including four homologous motifs, a connecting peptide, and N- and C-terminal extensions. β -crystallins constitute the major lens structural proteins, and they associate into dimers, tetramers and higher order aggregates. The β -crystallin subfamily is composed of several gene products, including β A1-, β A2-, β A3-, β A4-, β B1-, β B2- and β B3-crystallin. The β A1- and β A3-crystallin proteins are encoded by a single mRNA. They differ by only 17 amino acids, and β A1-crystallin is generated by use of an alternate translation initiation site.

REFERENCES

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2. Hejtmancik, J.F., et al. 1997. Association properties of β B2- and β A3-crystallin: ability to form dimers. *Protein Eng.* 10: 1347-1352.
3. Werten, P.J., et al. 1999. The short 5' untranslated region of the β A3/A1-crystallin mRNA is responsible for leaky ribosomal scanning. *Mol. Biol. Rep.* 26: 201-205.
4. Evans, P., et al. 2004. The P23T cataract mutation causes loss of solubility of folded γ D-crystallin. *J. Mol. Biol.* 343: 435-444.
5. Yang, Y., et al. 2004. Transcriptional regulation of mouse α B- and γ F-crystallin genes in lens: opposite promoter-specific interactions between Pax6 and large Maf transcription factors. *J. Mol. Biol.* 344: 351-368.
6. Gangalum, R.K., et al. 2004. Small heat shock protein α B-crystallin is part of cell cycle-dependent Golgi reorganization. *J. Biol. Chem.* 279: 43374-43377.
7. Kamradt, M.C., et al. 2005. The small heat shock protein α B-crystallin is a novel inhibitor of TRAIL-induced apoptosis that suppresses the activation of caspase-3. *J. Biol. Chem.* 280: 11059-11066.
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CHROMOSOMAL LOCATION

Genetic locus: CRYBB2 (human) mapping to 22q11.23.

PRODUCT

β B2-crystallin 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 β B2-crystallin shRNA Plasmid (h): sc-40444-SH and β B2-crystallin shRNA (h) Lentiviral Particles: sc-40444-V as alternate gene silencing products.

For independent verification of β B2-crystallin (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-40444A, sc-40444B and sc-40444C.

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

β B2-crystallin siRNA (h) is recommended for the inhibition of β B2-crystallin 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

β B2-crystallin (B-12): sc-376006 is recommended as a control antibody for monitoring of β B2-crystallin 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 β B2-crystallin gene expression knockdown using RT-PCR Primer: β B2-crystallin (h)-PR: sc-40444-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.