

# $\gamma$ B-crystallin siRNA (h): sc-40452

## BACKGROUND

Crystallins, the major proteins of the vertebrate eye lens, are responsible for maintaining the transparency and the refractive index of the lens. Crystallins are divided into  $\alpha$ ,  $\beta$ , and  $\gamma$  families, all of which usually contain seven distinctive protein regions, including four homologous motifs, one connecting peptide and N- and C-terminal extensions. The  $\gamma$ -crystallin family is comprised of seven closely related proteins designated  $\gamma$ A-,  $\gamma$ B-,  $\gamma$ C-,  $\gamma$ D-,  $\gamma$ E-,  $\gamma$ F- and  $\gamma$ G-crystallin.  $\gamma$ B-crystallin, also known as CRYGB or CRYG2, is a 175 amino acid member of the  $\gamma$ -crystallin family. Functioning as a monomer that has a two-domain  $\beta$  fold,  $\gamma$ B-crystallin, like other members of its family, plays a key role in ensuring the proper structure of the vertebrate eye lens. Defects in the gene encoding  $\gamma$ B-crystallin are associated with the formation of cataracts which are characterized by a clouding of the crystalline lens of the eye.

## REFERENCES

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3. Rogae, E.I., et al. 1996. Linkage of polymorphic congenital cataract to the  $\gamma$ -crystallin gene locus on human chromosome 2q33-35. *Hum. Mol. Genet.* 5: 699-703.
4. Graw, J. 1997. The crystallins: genes, proteins and diseases. *Biol. Chem.* 378: 1331-1348.
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6. Santhiya, S.T., et al. 2002. Novel mutations in the  $\gamma$ -crystallin genes cause autosomal dominant congenital cataracts. *J. Med. Genet.* 39: 352-358.
7. Salim, A., et al. 2003. Homology models of human  $\gamma$ -crystallins: structural study of the extensive charge network in  $\gamma$ -crystallins. *Biochem. Biophys. Res. Commun.* 300: 624-630.
8. Messina-Baas, O.M., et al. 2006. Two affected siblings with nuclear cataract associated with a novel missense mutation in the CRYGD gene. *Mol. Vis.* 12: 995-1000.

## CHROMOSOMAL LOCATION

Genetic locus: CRYGB (human) mapping to 2q34.

## PRODUCT

$\gamma$ B-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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see  $\gamma$ B-crystallin shRNA Plasmid (h): sc-40452-SH and  $\gamma$ B-crystallin shRNA (h) Lentiviral Particles: sc-40452-V as alternate gene silencing products.

For independent verification of  $\gamma$ B-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-40452A, sc-40452B and sc-40452C.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

$\gamma$ B-crystallin siRNA (h) is recommended for the inhibition of  $\gamma$ B-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  $\mu$ M in 66  $\mu$ 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

$\gamma$ B-crystallin (D-5): sc-377056 is recommended as a control antibody for monitoring of  $\gamma$ B-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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor  $\gamma$ B-crystallin gene expression knockdown using RT-PCR Primer:  $\gamma$ B-crystallin (h)-PR: sc-40452-PR (20  $\mu$ 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](http://www.scbt.com) for detailed protocols and support products.