



FOXE3 siRNA (h): sc-60653

BACKGROUND

The human forkhead-box (FOX) gene family consists of at least 43 members, including FOXE3, a 288 amino acid protein. FOXE3 is a winged-helix transcription factor that plays a crucial function during the initial stages of lens development and closure of the lens vesicle. FOXE3 may also act as a factor that promotes survival and proliferation, while preventing differentiation, in the lens epithelium. As the posterior cells of the lens fiber begin to differentiate, expression of FOXE3 is limited to the undifferentiated cells coating the anterior surface of the lens. Congenital primary aphakia (CPA) is a rare developmental disorder caused by a null mutation in the FOXE3 gene that is identified by the absence of a lens. The development of CPA is normally stimulated during the fourth or fifth week of human embryogenesis.

REFERENCES

1. Blixt, A., et al. 2000. A forkhead gene, FOXE3, is essential for lens epithelial proliferation and closure of the lens vesicle. *Genes Dev.* 14: 245-254.
2. Brownell, I., et al. 2000. Forkhead FOXE3 maps to the dysgenetic lens locus and is critical in lens development and differentiation. *Genesis* 27: 81-93.
3. Semina, E.V., et al. 2001. Mutations in the human forkhead transcription factor FOXE3 associated with anterior segment ocular dysgenesis and cataracts. *Hum. Mol. Genet.* 10: 231-236.
4. Lang, R.A. 2004. Pathways regulating lens induction in the mouse. *Int. J. Dev. Biol.* 48: 783-791.
5. Katoh, M. and Katoh, M. 2004. Human FOX gene family. *Int. J. Oncol.* 25: 1495-1500.
6. Yoshimoto, A., et al. 2005. Regulation of ocular lens development by Smad-interacting protein 1 involving FOXE3 activation. *Development* 132: 4437-4448.
7. Medina-Martinez, O., et al. 2005. Severe defects in proliferation and differentiation of lens cells in FOXE3 null mice. *Mol. Cell. Biol.* 25: 8854-8863.

CHROMOSOMAL LOCATION

Genetic locus: FOXE3 (human) mapping to 1p33.

PRODUCT

FOXE3 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 FOXE3 shRNA Plasmid (h): sc-60653-SH and FOXE3 shRNA (h) Lentiviral Particles: sc-60653-V as alternate gene silencing products.

For independent verification of FOXE3 (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-60653A, sc-60653B and sc-60653C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

FOXE3 siRNA (h) is recommended for the inhibition of FOXE3 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

FOXE3 (G-1): sc-393526 is recommended as a control antibody for monitoring of FOXE3 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 FOXE3 gene expression knockdown using RT-PCR Primer: FOXE3 (h)-PR: sc-60653-PR (20 μ l, 418 bp). 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.