

HESX1 siRNA (h): sc-38669

BACKGROUND

The homeobox protein, HESX1, which is also known as Rathke's pouch homeobox, HANF, homeodomain transcription factor, and anterior-restricted homeobox protein is a transcription factor that belongs to the homeodomain family of DNA binding proteins. HESX1 is initially expressed in embryonic stem cells and the primitive forebrain, and is essential for normal development of the eyes and other anterior CNS structures, such as the hypothalamus, the pituitary gland and the olfactory bulbs. The homeobox gene *Hesx1* is expressed in the anterior visceral endoderm (AVE), anterior axial mesendoderm (AME), and anterior neural ectoderm (ANE) during early embryogenesis. Mutations in the *Hesx1* gene are associated with disorders that are comparable with septo-optic dysplasia (SOD). These disorders are characterized by hypoplasia of the optic nerve, various types of forebrain defects and pituitary hormone deficiencies, including hypothyroidism. *Hesx1* also acts as a transcriptional repressor of reporter gene constructs in tissue culture assays.

REFERENCES

1. Dattani, M., et al. 1998. Mutations in the homeobox gene *HESX1/Hesx1* associated with septo-optic dysplasia in human and mouse. *Nat. Genet.* 19: 125-133.
2. Dattani, M., et al. 1999. *HESX1*: a novel gene implicated in a familial form of septo-optic dysplasia. *Acta Paediatr. Suppl.* 88: 49-54.
3. Dattani, M., et al. 2000. The molecular basis for developmental disorders of the pituitary gland in man. *Clin. Genet.* 57: 337-346.
4. Pfaffle, R., et al. 2000. Idiopathic growth hormone deficiency: a vanishing diagnosis? *Horm. Res.* 53: 1-8.
5. Dattani, M., et al. 2000. Molecular genetics of septo-optic dysplasia. *Horm. Res.* 53: 26-33.
6. Martinez-Barbera, J., et al. 2000. The homeobox gene *Hesx1* is required in the anterior neural ectoderm for normal forebrain formation. *Dev. Biol.* 223: 422-430.
7. Thomas, P., et al. 2001. Heterozygous *HESX1* mutations associated with isolated congenital pituitary hypoplasia and septo-optic dysplasia. *Hum. Mol. Genet.* 10: 39-45.

CHROMOSOMAL LOCATION

Genetic locus: *HESX1* (human) mapping to 3p14.3.

PRODUCT

HESX1 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 HESX1 shRNA Plasmid (h): sc-38669-SH and HESX1 shRNA (h) Lentiviral Particles: sc-38669-V as alternate gene silencing products.

For independent verification of HESX1 (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-38669A, sc-38669B and sc-38669C.

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

HESX1 siRNA (h) is recommended for the inhibition of HESX1 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

HESX1 (G-10): sc-515477 is recommended as a control antibody for monitoring of HESX1 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 HESX1 gene expression knockdown using RT-PCR Primer: HESX1 (h)-PR: sc-38669-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.