SANTA CRUZ BIOTECHNOLOGY, INC.

Six4 siRNA (h): sc-38790

BACKGROUND

The Six proteins (sine oculis) are a family of homeodomain transcription factors that share a conserved DNA binding domain. Six2, Six4 (AREC3) and Six5 bind to the same DNA sequence, indicating that they may regulate the same target genes. Six1 and Six4 are both capable of transactivating MEF3 site containing reporter genes, such as myogenin. It has been demonstrated that alterations to homeobox-containing genes may result in cancer. Six1 expression has been shown to be absent or low in normal adult tissues, although it is expressed in several tumor types, including breast carcinoma. Six1 overexpression has been shown to abrogate the G_2 cell cycle checkpoint.

REFERENCES

1. Kawakami, K., et al. 1996. Structure, function and expression of a murine homeobox protein AREC3, a homologue of *Drosophila* sine oculis gene product, and implication in development. Nucleic Acids Res. 24: 303-310.
2. Ohto, H., et al. 1998. Tissue and developmental distribution of Six family gene products. Int. J. Dev. Biol. 42: 141-148.
3. Ozaki, H., et al. 1999. Structure and chromosome mapping of the human SIX4 and murine Six4 genes. Cytogenet. Cell Genet. 87: 108-112.
4. Ozaki, H., et al. 2001. Six4, a putative myogenin gene regulator, is not essential for mouse embryonal development. Mol. Cell. Biol. 21: 3343-3350.
5. Ando, Z., et al. 2005. Slc12a2 is a direct target of two closely related homeobox proteins, Six1 and Six4. FEBS J. 272: 3026-3041.
6. Grifone, R., et al. 2005. Six1 and Six4 homeoproteins are required for Pax3 and Mrf expression during myogenesis in the mouse embryo. Development 132: 2235-2249.
7. Konishi, Y., et al. 2006. Six1 and Six4 promote survival of sensory neurons during early trigeminal gangliogenesis. Brain Res. 1116: 93-102.
8. Clark, I.B., et al. 2007. Live imaging of *Drosophila* gonad formation reveals roles for Six4 in regulating germline and somatic cell migration. BMC Dev. Biol. 7: 52.

CHROMOSOMAL LOCATION

Genetic locus: SIX4 (human) mapping to 14q23.1.

PRODUCT

Six4 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Six4 shRNA Plasmid (h): sc-38790-SH and Six4 shRNA (h) Lentiviral Particles: sc-38790-V as alternate gene silencing products.

For independent verification of Six4 (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-38790A, sc-38790B and sc-38790C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Six4 siRNA (h) is recommended for the inhibition of Six4 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 µM in 66 µl. 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

Six4 (D-5): sc-390779 is recommended as a control antibody for monitoring of Six4 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Six4 gene expression knockdown using RT-PCR Primer: Six4 (h)-PR: sc-38790-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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