SANTA CRUZ BIOTECHNOLOGY, INC.

Nkx-3.1 siRNA (m): sc-36078

BACKGROUND

The homeobox gene Nkx-3.1 is the human homolog of *Drosophila* bagpipe, which, in conjunction with tinman, determines cell fate in the dorsal mesoderm. In mammalian species, Nkx-3.1 is predominantly expressed in prostate, and it regulates prostate development in response to sonic hedgehog (Shh) signaling by exerting growth-suppressive and differentiating effects on prostatic epithelium. Nkx-3.1 is also expressed at lower levels in other tissues, including the heart and gut, in a Shh independent manner, where it plays a role in regulating proliferation of glandular epithelium and in the formation of ducts in prostate and minor salivary glands. Nkx-3.1 preferentially binds the TAAGTA sequence, which has not been reported for any other NK class homeoprotein. The human Nkx-3.1 gene is located on chromosome 8q21.2, which frequently undergoes a loss of heterozygosity, and although Nkx-3.1 is not a tumor suppressor gene, it may be a useful marker for benign and malignant prostate epithelium.

REFERENCES

1. Azpiazu, N., et al. 1993. tinman and bagpipe: two homeo box genes that determine cell fates in the dorsal mesoderm of *Drosophila*. Genes Dev. 7: 1325-1340.
2. Sciavolino, P.J., et al. 1997. Tissue-specific expression of murine Nkx-3.1 in the male urogenital system. Dev. Dyn. 209: 127-138.
3. Bowen, C., et al. 2000. Loss of Nkx-3.1 expression in human prostate cancers correlates with tumor progression. Cancer Res. 60: 6111-6115.
4. Schneider, A., et al. 2000. Targeted disruption of the Nkx-3.1 gene in mice results in morphogenetic defects of minor salivary glands: parallels to glandular duct morphogenesis in prostate. Mech. Dev. 95: 163-174.
5. Steadman, D.J., et al. 2000. DNA-binding sequence of the human prostate-specific homeodomain protein Nkx-3.1. Nucleic Acids Res. 28: 2389-2395.
6. Tanaka, M., et al. 2000. Nkx-3.1, a murine homolog of *Drosophila* bagpipe, regulates epithelial ductal branching and proliferation of the prostate and palatine glands. Dev. Dyn. 219: 248-260.

CHROMOSOMAL LOCATION

Genetic locus: Nkx3-1 (mouse) mapping to 14 D2.

PRODUCT

Nkx-3.1 siRNA (m) 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 Nkx-3.1 shRNA Plasmid (m): sc-36078-SH and Nkx-3.1 shRNA (m) Lentiviral Particles: sc-36078-V as alternate gene silencing products.

For independent verification of Nkx-3.1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-36078A, sc-36078B and sc-36078C.

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

Nkx-3.1 siRNA (m) is recommended for the inhibition of Nkx-3.1 expression in mouse 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

Nkx-3.1 (A-3): sc-393190 is recommended as a control antibody for monitoring of Nkx-3.1 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 Nkx-3.1 gene expression knockdown using RT-PCR Primer: Nkx-3.1 (m)-PR: sc-36078-PR (20 µl). 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.