SANTA CRUZ BIOTECHNOLOGY, INC.

frizzled-6 siRNA (h): sc-39987

BACKGROUND

Frizzled-6 (FZD6, also known as frizzled homolog 6 in *Drosophila*) is a seven-transmembrane domain receptor that binds the Wnt ligand and influences macroscopic hair patterning and other tissue polarity events. Frizzled-6 protein contains a signal peptide and a cysteine-rich domain (CRD) in the N-terminal extracellular region, and does not contain a C-terminal PDZ domain-binding motif. Frizzled-3 and frizzled-6 influence neural tube closure and the planar orientation of hair bundles on a subset of auditory and vestibular sensory cells. Madin-Darby canine kidney (MDCK) cells are competent to form tubules *in vitro* and express the frizzled-6 receptor, which is known to form a complex with Wnt-4 through the CRD in this cell type. Frizzled-6 is expressed as a 4.4 kb mRNA in various human tissues, including adult heart, brain, placenta, lung, liver, skeletal muscle, kidney, pancreas, thymus, prostate, testis, ovary, small intestine and colon.

REFERENCES

1. Tokuhara, M., et al. 1998. Molecular cloning of human frizzled-6. Biochem. Biophys. Res. Commun. 243: 622-627.
2. Yanagawa, S., et al. 1998. Identification and characterization of a novel line of *Drosophila* Schneider S2 cells that respond to wingless signaling. J. Biol. Chem. 273: 32353-32359.
3. Golan, T., et al. 2004. The human frizzled-6 (HFz6) acts as a negative regulator of the canonical Wnt. β-catenin signaling cascade. J. Biol. Chem. 279: 14879-14888.
4. Guo, N., et al. 2004. Frizzled-6 controls hair patterning in mice. Proc. Natl. Acad. Sci. USA 101: 9277-9281.
5. Lyons, J.P., et al. 2004. Wnt-4 activates the canonical β-catenin-mediated Wnt pathway and binds frizzled-6 CRD: functional implications of Wnt/β-catenin activity in kidney epithelial cells. Exp. Cell Res. 298: 369-387.
6. Gregorieff, A., et al. 2005. Expression pattern of Wnt signaling components in the adult intestine. Gastroenterology 129: 626-638.
7. Yang, L., et al. 2006. Bone morphogenetic protein-2 modulates Wnt and frizzled expression and enhances the canonical pathway of Wnt signaling in normal keratinocytes. J. Dermatol. Sci. 42: 111-119.

CHROMOSOMAL LOCATION

Genetic locus: FZD6 (human) mapping to 8q22.3.

PRODUCT

frizzled-6 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 frizzled-6 shRNA Plasmid (h): sc-39987-SH and frizzled-6 shRNA (h) Lentiviral Particles: sc-39987-V as alternate gene silencing products.

For independent verification of frizzled-6 (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-39987A, sc-39987B and sc-39987C.

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

frizzled-6 siRNA (h) is recommended for the inhibition of frizzled-6 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

frizzled-6 (C-12): sc-393791 is recommended as a control antibody for monitoring of frizzled-6 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 frizzled-6 gene expression knockdown using RT-PCR Primer: frizzled-6 (h)-PR: sc-39987-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.