

WIF-1 siRNA (h): sc-36837

BACKGROUND

The Wnt genes are a group of conserved, Cysteine-rich, secreted glycoproteins that are required for numerous developmental processes including embryogenesis, asymmetric cell division and central nervous system (CNS) patterning. Wnt association with the transmembrane spanning receptor frizzled activates dishevelled, which downregulates glycogen synthase kinase (GSK) through serine phosphorylation. Reduced levels of active GSK causes accumulation of β -catenin and subsequent regulation of developmentally significant Wnt target genes. Wnt antagonists such as dickkopf (Dkk), frizzled-related protein (sFRP) and Wnt inhibitory factor-1 (WIF-1), are necessary to ensure normal spatial and temporal patterns of Wnt activity during developmental processes. WIF-1 is a 379-amino acid secreted protein that contains an N-terminal signal sequence, a 150-amino acid WIF domain, five epidermal growth factor-like repeats and a 45-amino acid C-terminal hydrophilic domain.

REFERENCES

1. Krasnow, R.E., et al. 1995. Dishevelled is a component of the frizzled signaling pathway in *Drosophila*. *Development* 121: 4095-4102.
2. Cadigan, K.M., et al. 1997. Wnt signaling: a common theme in animal development. *Genes Dev.* 11: 3286-3305.
3. Sakanaka, C., et al. 1998. Bridging of β -catenin and glycogen synthase kinase-3 β by axin and inhibition of β -catenin-mediated transcription. *Proc. Natl. Acad. Sci. USA* 95: 3020-3023.
4. Glinka, A., et al. 1998. Dickkopf-1 is a member of a new family of secreted proteins and functions in head induction. *Nature* 391: 357-362.

CHROMOSOMAL LOCATION

Genetic locus: WIF1 (human) mapping to 12q14.3.

PRODUCT

WIF-1 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 WIF-1 shRNA Plasmid (h): sc-36837-SH and WIF-1 shRNA (h) Lentiviral Particles: sc-36837-V as alternate gene silencing products.

For independent verification of WIF-1 (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-36837A, sc-36837B and sc-36837C.

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

WIF-1 siRNA (h) is recommended for the inhibition of WIF-1 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

WIF-1 (B-10): sc-373780 is recommended as a control antibody for monitoring of WIF-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-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 WIF-1 gene expression knockdown using RT-PCR Primer: WIF-1 (h)-PR: sc-36837-PR (20 μ l, 431 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Urakami, S., et al. 2006. Epigenetic inactivation of Wnt inhibitory factor-1 plays an important role in bladder cancer through aberrant canonical Wnt/ β -catenin signaling pathway. *Clin. Cancer Res.* 12: 383-391.
2. Xu, J.H., et al. 2015. Role of Wnt inhibitory factor-1 in inhibition of bisdemethoxycurcumin mediated epithelial-to-mesenchymal transition in highly metastatic lung cancer 95D cells. *Chin. Med. J.* 128: 1376-1383.

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.