

Kremen-1 siRNA (h): sc-60899

BACKGROUND

The Wnt genes are a group of well conserved, cysteine-rich secreted glycoproteins that are required for numerous developmental processes including embryogenesis, asymmetric cell division and central nervous system (CNS) patterning. The association of the Wnt protein with the seven membrane spanning receptor frizzled activates dishevelled, which downregulates glycogen synthase kinase (GSK) through serine phosphorylation. Reduced levels of active GSK causes the accumulation of β -catenin and subsequent regulation of developmentally significant Wnt target genes. Wnt antagonists, Dickkopf (which includes Dkk1-4), frizzled-related protein (sFRP), Soggy-1, Kremen-1 and Wnt inhibitory factor-1 (WIF-1) are necessary to ensure normal spatial and temporal patterns of Wnt activity during developmental processes.

REFERENCES

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2. Cadigan, K.M. and Nusse, R. 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.
5. Fedi, P., et al. 1999. Isolation and biochemical characterization of the human Dkk-1 homologue, a novel inhibitor of mammalian Wnt signaling. *J. Biol. Chem.* 274: 19465-19472.
6. Etheridge, S.L., et al. 2004. Expression profiling and functional analysis of Wnt signaling mechanisms in mesenchymal stem cells. *Stem Cells* 22: 849-860.
7. Kulkarni, N.H., et al. 2005. Effects of parathyroid hormone on Wnt signaling pathway in bone. *J. Cell. Biochem.* 95: 1178-1190.

CHROMOSOMAL LOCATION

Genetic locus: KREMEN1 (human) mapping to 22q12.1.

PRODUCT

Kremen-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 Kremen-1 shRNA Plasmid (h): sc-60899-SH and Kremen-1 shRNA (h) Lentiviral Particles: sc-60899-V as alternate gene silencing products.

For independent verification of Kremen-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-60899A, sc-60899B and sc-60899C.

PROTOCOLS

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

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

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

Kremen-1 (R17-2): sc-74206 is recommended as a control antibody for monitoring of Kremen-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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Kremen-1 gene expression knockdown using RT-PCR Primer: Kremen-1 (h)-PR: sc-60899-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.