SANTA CRUZ BIOTECHNOLOGY, INC.

gremlin-1 siRNA (h): sc-39408

BACKGROUND

The gremlin protein family contains antagonists of bone morphogenetic protein (BMP) signaling that are expressed in the neural crest. All family members are secreted proteins that act as BMP antagonists in embryonic explants and are expressed in the proximal airway epithelium of the lung during embryonic development. Gremlin-1 is required for early limb outgrowth and patterning in the FGF4-SHH feedback loop, while gremlin-2 binds and blocks the activity of BMP-2 and BMP-4. The dorsaling factor gremlin belongs to a novel gene family that includes the head-inducing factor Cerberus and the tumor suppressor DAN. Additionally, secreted gremlin relays the Sonic hedgehog signal from the polarizing region to the apical ectodermal ridge.

REFERENCES

1. Hsu, D.R., et al. 1998. The *Xenopus* dorsalizing factor gremlin identifies a novel family of secreted proteins that antagonize BMP activities. Mol. Cell 1: 673-683.
2. Zuniga, A., et al. 1999. Signal relay by BMP antagonism controls the Shh/FGF4 feedback loop in vertebrate limb buds. Nature 401: 598-602.
3. Topol, L.Z., et al. 2000. DRM/gremlin (CKTSF1B1) maps to human chromosome 15 and is highly expressed in adult and fetal brain. Cytogenet. Cell Genet. 89: 79-84.
4. Ohtori, S., et al. 2001. A novel neurotransmitter, DAN, mediates pain sensation in the spinal dorsal horn. Nippon Rinsho 59: 1698-1703.
5. Shi, W., et al. 2001. Gremlin negatively modulates BMP-4 induction of embryonic mouse lung branching morphogenesis. Am. J. Physiol. Lung Cell. Mol. Physiol. 280: 1030-1039.
6. Lu, M.M., et al. 2001. The bone morphogenic protein antagonist gremlin regulates proximal-distal patterning of the lung. Dev. Dyn. 222: 667-680.

CHROMOSOMAL LOCATION

Genetic locus: GREM1 (human) mapping to 15q13.3.

PRODUCT

gremlin-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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see gremlin-1 shRNA Plasmid (h): sc-39408-SH and gremlin-1 shRNA (h) Lentiviral Particles: sc-39408-V as alternate gene silencing products.

For independent verification of gremlin-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-39408A, sc-39408B and sc-39408C.

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.

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

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

gremlin-1 (C-7): sc-515877 is recommended as a control antibody for monitoring of gremlin-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 gremlin-1 gene expression knockdown using RT-PCR Primer: gremlin-1 (h)-PR: sc-39408-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.