

Sec5 siRNA (h): sc-106891

BACKGROUND

The mammalian exocyst complex (Sec 6/8) is a multiple protein complex essential for targeting exocytic vesicles to specific docking sites on the plasma membrane. Sec5 is one of eight exocyst complex subunits: Sec3, Sec5, Sec6, Sec8, Sec10, Sec15, Exo70 and Exo84. Exocyst complex regulation depends upon Sec5 binding Ral in a GTP-dependent manner. Human Sec5 maps to chromosome 6p25.3.

REFERENCES

1. Fukui, S., et al. 2003. Structural basis of the interaction between Ral A and Sec5, a subunit of the Sec6/8 complex. *EMBO J.* 13: 3267-3278.
2. Mott, H.R., et al. 2003. Structure of the GTPase-binding domain of Sec5 and elucidation of its Ral binding site. *J. Biol. Chem.* 278: 17053-17059.
3. Moskalenko, S., et al. 2003. Ral GTPases regulate exocyst assembly through dual subunit interactions. *J. Biol. Chem.* 278: 51743-51748.
4. Murthy, M., et al. 2003. Mutations in the exocyst component Sec5 disrupt neuronal membrane traffic, but neurotransmitter release persists. *Neuron* 37: 433-447.
5. Murthy, M., et al. 2004. The exocyst component Sec5 is required for membrane traffic and polarity in the *Drosophila* ovary. *Development* 131: 377-388.
6. Wang, S., et al. 2004. The mammalian exocyst, a complex required for exocytosis, inhibits tubulin polymerization. *J. Biol. Chem.* 279: 35958-35966.
7. LocusLink Report (LocusID: 55770). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: EXOC2 (human) mapping to 6p25.3.

PRODUCT

Sec5 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 Sec5 shRNA Plasmid (h): sc-106891-SH and Sec5 shRNA (h) Lentiviral Particles: sc-106891-V as alternate gene silencing products.

For independent verification of Sec5 (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-106891A, sc-106891B and sc-106891C.

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

Sec5 siRNA (h) is recommended for the inhibition of Sec5 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

Sec5 (F-7): sc-393230 is recommended as a control antibody for monitoring of Sec5 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 Sec5 gene expression knockdown using RT-PCR Primer: Sec5 (h)-PR: sc-106891-PR (20 μ l, 560 bp). 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.