SANTA CRUZ BIOTECHNOLOGY, INC.

Exo84 siRNA (m): sc-144970

BACKGROUND

Exocytosis is crucial in membrane trafficking and it mediates hormone and neurotransmitter secretion out of the cell, as well as the incorporation of membrane proteins and lipids to the plasma membrane. It is crucial for cell-cell communication, cell growth and cell polarity. The exocyst complex is a multiprotein complex that consists of Sec3, Sec5, Sec6, Sec8, Sec10, Sec15, Exo70 and Exo84, and is essential for targeting exocytic vesicles to specific docking sites on the plasma membrane. The exocyst complex inhibits tubulin polymerization *in vitro,* suggesting that the exocyst complex is important for modulating the microtubule dynamics that underlie exocytosis. Exo84 (exocyst complex 84 kDa subunit), also known as Exocyst complex component 8, is a 725 amino acid protein that is one of eight protein subunits composing the mammalian Exocyst complex. Both Exo84 and Sec5 are effector targets for active Ral GTPases, which are responsible for regulating exocyst complex activities.

REFERENCES

1. Guo, W., et al. 1999. Exo84p is an exocyst protein essential for secretion. J. Biol. Chem. 274: 23558-23564.
2. Moskalenko, S., et al. 2003. Ral GTPases regulate exocyst assembly through dual subunit interactions. J. Biol. Chem. 278: 51743-51748.
3. Hsu, S.C., et al. 2004. The exocyst complex in polarized exocytosis. Int. Rev. Cytol. 233: 243-265.
4. Wang, S., et al. 2004. The mammalian exocyst, a complex required for exocytosis, inhibits tubulin polymerization. J. Biol. Chem. 279: 35958-35966.
5. Jin, R., et al. 2005. Exo84 and Sec5 are competitive regulatory Sec6/8 effectors to the RalA GTPase. EMBO J. 24: 2064-2074.
6. Dong, G., et al. 2005. The structures of exocyst subunit Exo70p and the Exo84p C-terminal domains reveal a common motif. Nat. Struct. Mol. Biol. 12: 1094-1100.
7. Lalli, G. 2009. RalA and the exocyst complex influence neuronal polarity through PAR-3 and aPKC. J. Cell Sci. 122: 1499-1506.
8. Issaq, S.H., et al. 2010. Sec5 and exo84 foster oncogenic Ras-mediated tumorigenesis. Mol. Cancer Res. 8: 223-231.

CHROMOSOMAL LOCATION

Genetic locus: Exoc8 (mouse) mapping to 8 E2.

PRODUCT

Exo84 siRNA (m) is a pool of 2 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 Exo84 shRNA Plasmid (m): sc-144970-SH and Exo84 shRNA (m) Lentiviral Particles: sc-144970-V as alternate gene silencing products.

For independent verification of Exo84 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-144970A and sc-144970B.

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

Exo84 siRNA (m) is recommended for the inhibition of Exo84 expression in mouse 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

Exo84 (H-1): sc-515532 is recommended as a control antibody for monitoring of Exo84 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 Exo84 gene expression knockdown using RT-PCR Primer: Exo84 (m)-PR: sc-144970-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.