SANTA CRUZ BIOTECHNOLOGY, INC.

SNAP 23 siRNA (h): sc-41308

BACKGROUND

In eukaryotic cells, the Golgi apparatus receives newly synthesized proteins from the endoplasmic reticulum and delivers them after covalent modification to their destination in the cell. For membrane-directed proteins this process is believed to be carried out via vesicular transport. Correct vesicular transport is determined by specific pairing of vesicle-associated SNAREs (v-SNAREs) with those on the target membrane (t-SNAREs). This complex then recruits soluble NSF attachment proteins (SNAPs) and N-ethylmaleimide-sensitive factor (NSF) to form the highly stable SNAP receptor (SNARE) complex. The formation of a SNARE complex pulls the vesicle and target membrane together and may provide the energy to drive fusion of the lipid bilayers. A SNAP 25 related t-SNARE protein, SNAP 23, is required for exocytosis, suggesting that SNAP 23 may play an important role in membrane fusion events. The human SNAP 23 gene encodes two SNAP 23 isoforms, SNAP 23A and SNAP 23B. SNAP 23B is identical to a fragment of SNAP 23A, but SNAP 23B lacks 53 amino acid residues (90 to 142) that are present in SNAP 23A. SNAP 23 is ubiquitously expressed and is an important regulator of transport vesicle docking and fusion in all mammalian cells.

REFERENCES

1. Nagahama, M., et al. 1996. A v-SNARE implicated in intra-Golgi transport. J. Cell Biol. 133: 507-516.
2. Ravichandran, V., et al. 1996. Identification of a novel syntaxin- and synaptobrevin/VAMP-binding protein, SNAP 23, expressed in non-neuronal tissues. J. Biol. Chem. 271: 13300-13333.
3. Lowe, S.L., et al. 1997. A SNARE involved in protein transport through the Golgi apparatus. Nature 389: 881-884.
4. Mollinedo, F., et al. 1997. Identification of two isoforms of the vesicle-membrane fusion protein SNAP 23 in human neutrophils and HL-60 cells. Biochem. Biophys. Res. Commun. 231: 808-812.
5. Guo, Z., et al. 1998. Relocation of the t-SNARE SNAP 23 from lamellipodia-like cell surface projections regulates compound exocytosis in mast cells. Cell 94: 537-548.
6. Bentz, J. and Mittal, A. 2000. Deployment of membrane fusion protein domains during fusion. Cell Biol. Int. 24: 819-838.

CHROMOSOMAL LOCATION

Genetic locus: SNAP23 (human) mapping to 15q15.1.

PRODUCT

SNAP 23 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 SNAP 23 shRNA Plasmid (h): sc-41308-SH and SNAP 23 shRNA (h) Lentiviral Particles: sc-41308-V as alternate gene silencing products.

For independent verification of SNAP 23 (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-41308A, sc-41308B and sc-41308C.

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

SNAP 23 siRNA (h) is recommended for the inhibition of SNAP 23 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

SNAP 23 (D-11): sc-374215 is recommended as a control antibody for monitoring of SNAP 23 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 SNAP 23 gene expression knockdown using RT-PCR Primer: SNAP 23 (h)-PR: sc-41308-PR (20 µl, 541 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.