

α-SNAP siRNA (h): sc-29617

BACKGROUND

Syntaxins were originally thought to be docking proteins, but have more recently been categorized as anchoring proteins that anchor themselves to the cytoplasmic surfaces of cellular membranes. Syntaxins have been shown to bind to various proteins involved in exocytosis, including VAMPs (vesicle-associated membrane proteins), NSF (N-ethylmaleimide-sensitive factor), SNAP 25 (synaptosomal-associated protein of 25 kDa), SNAPs (soluble NSF attachment proteins) and synaptotagmin. VAMPs, also designated synaptobrevins, including VAMP-1 and VAMP-2, and synaptotagmin, a protein that may function as an inhibitor of exocytosis, are vesicular proteins. SNAPs, including α- and γ-SNAP, are cytoplasmic proteins that bind to a membrane receptor complex composed of VAMP, SNAP 25 and Syntaxin. SNAPs mediate the membrane binding of NSF, which is essential for membrane fusion reactions. An additional protein designated synaptophysin may regulate exocytosis by competing with SNAP 25 and Syntaxins for VAMP binding.

REFERENCES

1. Bachem, C.W., et al. 2000. Antisense suppression of a potato α-SNAP homologue leads to alterations in cellular development and assimilate distribution. *Plant Mol. Biol.* 43: 473-482.
2. Graham, M.E. and Burgoyne, R.D. 2000. Comparison of cysteine string protein (CSP) and mutant α-SNAP overexpression reveals a role for CSP in late steps of membrane fusion in dense-core granule exocytosis in adrenal chromaffin cells. *J. Neurosci.* 20: 1281-1289.
3. Wang, L., et al. 2000. The docking of primed vacuoles can be reversibly arrested by excess Sec17p (α-SNAP). *J. Biol. Chem.* 275: 22862-22867.
4. Scales, S.J., et al. 2001. The ionic layer is required for efficient dissociation of the SNARE complex by α-SNAP and NSF. *Proc. Natl. Acad. Sci. USA* 98: 14262-14267.
5. Marz, K.E., et al. 2003. Defining the SNARE complex binding surface of α-SNAP: implications for SNARE complex disassembly. *J. Biol. Chem.* 278: 27000-27008.
6. Chae, T.H., et al. 2004. The hyh mutation uncovers roles for α-SNAP in apical protein localization and control of neural cell fate. *Nat. Genet.* 36: 264-270.

CHROMOSOMAL LOCATION

Genetic locus: NAPA (human) mapping to 19q13.32.

PRODUCT

α-SNAP 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 α-SNAP shRNA Plasmid (h): sc-29617-SH and α-SNAP shRNA (h) Lentiviral Particles: sc-29617-V as alternate gene silencing products.

For independent verification of α-SNAP (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-29617A, sc-29617B and sc-29617C.

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

α-SNAP siRNA (h) is recommended for the inhibition of α-SNAP 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor α-SNAP gene expression knockdown using RT-PCR Primer: α-SNAP (h)-PR: sc-29617-PR (20 μl, 544 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Andreeva, A.V., et al. 2005. G_α12 interaction with alphaSNAP induces VE-cadherin localization at endothelial junctions and regulates barrier function. *J. Biol. Chem.* 280: 30376-30383.
2. Lee, J.E., et al. 2012. Nongenomic Stat5-dependent effects on Golgi apparatus and endoplasmic reticulum structure and function. *Am. J. Physiol., Cell Physiol.* 302: C804-C820.

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.