

SNAP 25 siRNA (h): sc-36517

BACKGROUND

Syntaxins were originally thought to be docking proteins but have now 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, 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

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4. Hayashi, T., et al. 1994. Synaptic vesicle membrane fusion complex: action of clostridial neurotoxins on assembly. *EMBO J.* 13: 5051-5061.
5. Edelman, L., et al. 1995. Synaptobrevin binding to synaptophysin: a potential mechanism for controlling the exocytosis fusion machine. *EMBO J.* 14: 224-231.
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CHROMOSOMAL LOCATION

Genetic locus: SNAP25 (human) mapping to 20p12.2.

PRODUCT

SNAP 25 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 SNAP 25 shRNA Plasmid (h): sc-36517-SH and SNAP 25 shRNA (h) Lentiviral Particles: sc-36517-V as alternate gene silencing products.

For independent verification of SNAP 25 (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-36517A, sc-36517B and sc-36517C.

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

SNAP 25 siRNA (h) is recommended for the inhibition of SNAP 25 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

SNAP 25 (SP12): sc-20038 is recommended as a control antibody for monitoring of SNAP 25 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 SNAP 25 gene expression knockdown using RT-PCR Primer: SNAP 25 (h)-PR: sc-36517-PR (20 μ l, 495 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.