

SNX5 siRNA (m): sc-41356

BACKGROUND

Sorting nexin 1 (SNX1) is a member of a large family of hydrophilic proteins that interact with a variety of receptor types and are involved in intracellular trafficking. SNX1 and the related splice variant, SNX1A, bind the epidermal growth factor (EGF) receptor, facilitate its transport to lysosome, and thereby contribute to the degradation of the receptor. SNX2 and SNX4 share a high degree of amino acid similarity with SNX1, as they all contain a characteristic phox homology (PX) domain. These proteins are all partially associated with cellular membranes, and they, likewise, associate with EGF, PDGF and Insulin receptor tyrosine kinases. These nexins are widely expressed and yet have various tissue distribution patterns. Additionally, the sorting nexins can associate with each other and with a variety of other cellular proteins, suggesting that they exist as part of multisubunit complexes. The related protein, SNX3, comprises a distinct subgroup of nexins that share less sequence similarity outside of the PX domain and have dramatically different binding affinities for the tyrosine kinase receptors.

REFERENCES

1. Trowbridge, I.S., et al. 1993. Signal-dependent membrane protein trafficking in the endocytic pathway. *Annu. Rev. Cell Biol.* 9: 129-161.
2. Opresko, L.K., et al. 1995. Endocytosis and lysosomal targeting of epidermal growth factor receptors are mediated by distinct sequences independent of the tyrosine kinase domain. *J. Biol. Chem.* 270: 4325-4333.
3. Ponting, C.P. 1996. Novel domains in NADPH oxidase subunits, sorting nexins, and PtdIns 3-kinases: binding partners of SH3 domains? *Protein Sci.* 5: 2353-2357.
4. Kurten, R.C., et al. 1996. Enhanced degradation of EGF receptors by a sorting nexin, SNX1. *Science* 272: 1008-1010.
5. Horadzovsky, B.F., et al. 1997. A sorting nexin-1 homologue, Vps5p, forms a complex with Vps17p and is required for recycling the vacuolar protein-sorting receptor. *Mol. Biol. Cell* 8: 1529-1541.
6. Haft, C.R., et al. 1998. Identification of a family of sorting nexin molecules and characterization of their association with receptors. *Mol. Cell. Biol.* 18: 7278-7287.

CHROMOSOMAL LOCATION

Genetic locus: Snx5 (mouse) mapping to 2 G1.

PRODUCT

SNX5 siRNA (m) 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 SNX5 shRNA Plasmid (m): sc-41356-SH and SNX5 shRNA (m) Lentiviral Particles: sc-41356-V as alternate gene silencing products.

For independent verification of SNX5 (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-41356A, sc-41356B and sc-41356C.

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

SNX5 siRNA (m) is recommended for the inhibition of SNX5 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 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

SNX5 (F-11): sc-515215 is recommended as a control antibody for monitoring of SNX5 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 SNX5 gene expression knockdown using RT-PCR Primer: SNX5 (m)-PR: sc-41356-PR (20 μ l, 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.