

SNX16 siRNA (h): sc-61585

BACKGROUND

Sorting nexin (SNX) proteins are members of a large family of hydrophilic proteins that interact with a variety of receptor types, contain a characteristic phox homology (PX) domain and play a role in intracellular trafficking. Viral infection by a nucleocapsid is initiated when it is transported to late endosomes and, ultimately, to the cytoplasm. This process depends on the late endosomal lipid lysobisphosphatidic acid (LBPA) and its putative effector Alix/AIP1, and is regulated by PtdIns3P (phosphatidylinositol-3-phosphate) signaling via SNX16, its binding protein. Overexpression of SNX16 increases the rate of EGF-induced EGFR degradation and prevents EGF-induced upmodulation of ERK and serum response element (SRE). Mutation in the PX domain eradicates the inhibitory effect of SNX16 on EGF-induced activation of ERK and SRE, suggesting that SNX16 directs the sorting of EGFR to the endosomal compartment, thus regulating EGF-induced cell signaling.

REFERENCES

1. Hanson, B.J., et al. 2003. Evidence for a role of SNX16 in regulating traffic between the early and later endosomal compartments. *J. Biol. Chem.* 278: 34617-34630.
2. Choi, J.H., et al. 2004. Sorting nexin 16 regulates EGF receptor trafficking by phosphatidylinositol-3-phosphate interaction with the phox domain. *J. Cell Sci.* 117: 4209-4218.
3. Watahiki, A., et al. 2004. Libraries enriched for alternatively spliced exons reveal splicing patterns in melanocytes and melanomas. *Nat. Methods* 1: 233-239.
4. Le Blanc, I., et al. 2005. Endosome-to-cytosol transport of viral nucleocapsids. *Nat. Cell Biol.* 7: 653-664.

CHROMOSOMAL LOCATION

Genetic locus: SNX16 (human) mapping to 8q21.13.

PRODUCT

SNX16 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 SNX16 shRNA Plasmid (h): sc-61585-SH and SNX16 shRNA (h) Lentiviral Particles: sc-61585-V as alternate gene silencing products.

For independent verification of SNX16 (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-61585A, sc-61585B and sc-61585C.

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.

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

SNX16 siRNA (h) is recommended for the inhibition of SNX16 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

SNX16 (C-11): sc-271260 is recommended as a control antibody for monitoring of SNX16 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 SNX16 gene expression knockdown using RT-PCR Primer: SNX16 (h)-PR: sc-61585-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.