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▶ Snapin siRNA (m): sc-45546

BACKGROUND

Biogenesis of lysosome-related organelles complex-1 (BLOC-1) is a multi-subunit protein necessary for biogenesis of specialized organelles of the endosomal-lysosomal system (such as melanosomes and platelet-dense granules). The complex consists of coiled-coil-forming proteins Snapin, Pallidin, Cappuccino, Muted, BLOS1, BLOS2 and BLOS3. The localization of these proteins varies as they can be cytoplasmic, peripheral membrane bound or anchored to the vesicular membrane. Snapin associates with the SNARE complex in neurons. Phosphorylation of Snapin increases its interaction with SNAP25 and affects exocytotic protein interactions. Snapin interacts with the N-terminus of regulator of G protein signaling 7 (RGS7), which is important in synaptic vesicle exocytosis.

REFERENCES

1. Ilardi, J.M., et al. 1999. Snapin: a SNARE-associated protein implicated in synaptic transmission. *Nat. Neurosci.* 2: 119-124.
2. Hunt, R.A., et al. 2003. Snapin interacts with the N-terminus of regulator of G protein signaling 7. *Biochem. Biophys. Res. Commun.* 303: 594-599.
3. Buxton, P., et al. 2003. Identification and characterization of Snapin as a ubiquitously expressed SNARE-binding protein that interacts with SNAP23 in non-neuronal cells. *Biochem. J.* 375: 433-440.
4. Starcevic, M., et al. 2004. Identification of Snapin and three novel proteins (BLOS1, BLOS2, and BLOS3/reduced pigmentation) as subunits of biogenesis of lysosome-related organelles complex-1 (BLOC-1). *J. Biol. Chem.* 279: 28393-28401.
5. Chou, J.L., et al. 2004. Regulation of type VI adenylyl cyclase by Snapin, a SNAP25-binding protein. *J. Biol. Chem.* 279: 46271-46279.

CHROMOSOMAL LOCATION

Genetic locus: Snapin (mouse) mapping to 3 F1.

PRODUCT

Snapin 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 Snapin shRNA Plasmid (m): sc-45546-SH and Snapin shRNA (m) Lentiviral Particles: sc-45546-V as alternate gene silencing products.

For independent verification of Snapin (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-45546A, sc-45546B and sc-45546C.

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

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

Snapin (E-10): sc-514675 is recommended as a control antibody for monitoring of Snapin 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 Snapin gene expression knockdown using RT-PCR Primer: Snapin (m)-PR: sc-45546-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.