

Unc18-1 siRNA (r): sc-270657

BACKGROUND

Unc18-1, 2 and 3 (syntaxin binding proteins 1-3, STXBP1-3, UNC18-a-c, MUNC18-1-3) are chaperone molecules that block syntaxin interactions with cognate SNARE (soluble NSF attachment protein (SNAP) receptors) proteins and regulate exocytosis. Unc18-1-3 mRNA is present in RBL-2H3 mast cells, mouse bone marrow derived mast cells (BMMC), and platelets. Unc18-1 Ser 313 is a protein kinase C phosphorylation site and Thr 574 is a cyclin-dependent kinase 5 phosphorylation site that regulates Unc18-1/Syntaxin1A interactions. Unc18-1 is phosphorylated on Ser 313 in response to phorbol ester treatment in adrenal chromaffin cells. Unc18-2 co-localizes with Syntaxin 3 at the apical plasma membrane of intestinal, proximal tubule and collecting duct epithelial cells.

REFERENCES

- Schraw, T.D., et al. 2003. A role for Sec1/Munc18 proteins in platelet exocytosis. *Biochem. J.* 374: 207-217.
- Barclay, J.W., et al. 2003. Phosphorylation of Munc18 by protein kinase C regulates the kinetics of exocytosis. *J. Biol. Chem.* 278: 10538-10545.
- Gaisano, H.Y., et al. 2004. Alcoholic chronic pancreatitis involves displacement of Munc18c from the pancreatic acinar basal membrane surface. *Pancreas* 28: 395-400.
- Graham, M.E., et al. 2004. Syntaxin/Munc18 interactions in the late events during vesicle fusion and release in exocytosis. *J. Biol. Chem.* 279: 32751-32760.
- Gladychева, S.E., et al. 2004. Regulation of syntaxin1A-munc18 complex for SNARE pairing in HEK293 cells. *J. Physiol.* 558: 857-871.
- Liu, J., et al. 2004. Fluorescence resonance energy transfer reports properties of syntaxin1a interaction with Munc18-1 *in vivo*. *J. Biol. Chem.* 279: 55924-55936.
- Chiufò, L.F., et al. 2005. Munc18-1 regulates early and late stages of exocytosis via syntaxin-independent protein interactions. *Mol. Biol. Cell* 16: 470-482.
- Nigam, R., et al. 2005. Expression and transcriptional regulation of Munc18 isoforms in mast cells. *Biochim. Biophys. Acta* 1728: 77-83.

CHROMOSOMAL LOCATION

Genetic locus: Stxbp1 (rat) mapping to 3p11.

PRODUCT

Unc18-1 siRNA (r) 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 Unc18-1 shRNA Plasmid (r): sc-270657-SH and Unc18-1 shRNA (r) Lentiviral Particles: sc-270657-V as alternate gene silencing products.

For independent verification of Unc18-1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270657A, sc-270657B and sc-270657C.

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

Unc18-1 siRNA (r) is recommended for the inhibition of Unc18-1 expression in rat 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

Unc18-1 (31): sc-136304 is recommended as a control antibody for monitoring of Unc18-1 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Unc18-1 gene expression knockdown using RT-PCR Primer: Unc18-1 (r)-PR: sc-270657-PR (20 μ l, 568 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.