

Sec63 siRNA (m): sc-41289

BACKGROUND

In mammalian cells, protein translocation across the endoplasmic reticulum (ER) membrane is almost exclusively co-translational. This transport depends on the Sec61 complex, which is homologous to the yeast Sec61p complex and has been identified in mammals as a ribosome-bound pore-forming membrane protein complex. The Sec61 complex associates with two ubiquitous ER membrane proteins Sec62 (also designated human translocation protein 1 or HTP1) and Sec63. The Sec61 complex forms the hydrophilic pore in the membrane through which the nascent polypeptide is translocated. Sec61p seems to be the evolutionary conserved component since homologues of Sec61p have been found both in bacteria and mammals. Sec62 is expressed in various human tissues such as the heart, brain, placenta, liver and pancreas.

REFERENCES

1. Simon, S.M. and Blobel, G. 1991. A protein-conducting channel in the endoplasmic reticulum. *Cell* 65: 371-380.
2. Görlich, D. and Rapoport, T.A. 1993. Protein translocation into proteoliposomes reconstituted from purified components of the endoplasmic reticulum membrane. *Cell* 75: 615-630.
3. Rapoport, T.A., et al. 1996. Protein transport across the eukaryotic endoplasmic reticulum and bacterial inner membranes. *Annu. Rev. Biochem.* 65: 271-303.
4. Hanein, D., et al. 1996. Oligomeric rings of the Sec61p complex induced by ligands required for protein translocation. *Cell* 87: 721-732.
5. Daimon, M., et al. 1997. Identification of a human cDNA homologue to the *Drosophila* translocation protein 1 (Dtrp1). *Biochem. Biophys. Res. Commun.* 230: 100-104.
6. Beckmann, R., et al. 1997. Alignment of conduits for the nascent polypeptide chain in the ribosome-Sec61 complex. *Science* 278: 2123-2126.
7. Meyer, H.A., et al. 2000. Mammalian Sec61 is associated with Sec62 and Sec63. *J. Biol. Chem.* 275: 14550-14557.

CHROMOSOMAL LOCATION

Genetic locus: Sec63 (mouse) mapping to 10 B2.

PRODUCT

Sec63 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 Sec63 shRNA Plasmid (m): sc-41289-SH and Sec63 shRNA (m) Lentiviral Particles: sc-41289-V as alternate gene silencing products.

For independent verification of Sec63 (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-41289A, sc-41289B and sc-41289C.

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

Sec63 siRNA (m) is recommended for the inhibition of Sec63 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

Sec63 (1A8): sc-517139 is recommended as a control antibody for monitoring of Sec63 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 Sec63 gene expression knockdown using RT-PCR Primer: Sec63 (m)-PR: sc-41289-PR (20 μ l). 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.