



ALG10B siRNA (m): sc-141010

BACKGROUND

Glycosylation of asparagine residues is an essential protein modification reaction that occurs upon most proteins that enter the secretory pathway in eukaryotic cells. Asparagine-linked oligosaccharides are transferred onto polypeptides in the lumen of the rough endoplasmic reticulum. ALG10/ALG10B, also known as DIE2 or KCR1, is a 473 amino acid multi-pass membrane protein that localizes to the endoplasmic reticulum. ALG10/ALG10B adds the third glucose residue to the lipid-linked oligosaccharide precursor for N-linked glycosylation and transfers glucose from dolichyl phosphate glucose onto the lipid-linked Glc2Man9GlcNAc2 oligosaccharide.

REFERENCES

1. Burda, P. and Aebi, M. 1998. The ALG10 locus of *Saccharomyces cerevisiae* encodes the α -1,2 glucosyltransferase of the endoplasmic reticulum: the terminal glucose of the lipid-linked oligosaccharide is required for efficient N-linked glycosylation. *Glycobiology* 8: 455-462.
2. de L Ufret, M. and Imperiali, B. 2000. Probing the extended binding determinants of oligosaccharyl transferase with synthetic inhibitors of asparagine-linked glycosylation. *Bioorg. Med. Chem. Lett.* 10: 281-284.
3. Nagayama, Y., et al. 2000. Identification of the sites of asparagine-linked glycosylation on the human thyrotropin receptor and studies on their role in receptor function and expression. *J. Pharmacol. Exp. Ther.* 295: 404-409.
4. Uchimura, S., et al. 2005. Effects of N-glycosylation and inositol on the ER stress response in yeast *Saccharomyces cerevisiae*. *Biosci. Biotechnol. Biochem.* 69: 1274-1280.
5. Robbins, P.W. and Samuelson, J. 2005. Asparagine linked glycosylation in *Giardia*. *Glycobiology* 15: 15G-16G.
6. Nakajima, T., et al. 2007. HERG is protected from pharmacological block by α -1,2-glucosyltransferase function. *J. Biol. Chem.* 282: 5506-5513.
7. Drago, A., et al. 2008. Strategy for a genetic assessment of antipsychotic and antidepressant-related proarrhythmia. *Curr. Med. Chem.* 15: 2472-2517.
8. Grabinska, K.A., et al. 2008. Dolichyl-phosphate-glucose is used to make O-glycans on glycoproteins of *Trichomonas vaginalis*. *Eukaryotic Cell* 7: 1344-1351.

CHROMOSOMAL LOCATION

Genetic locus: Alg10b (mouse) mapping to 15 E3.

PRODUCT

ALG10B 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 ALG10B shRNA Plasmid (m): sc-141010-SH and ALG10B shRNA (m) Lentiviral Particles: sc-141010-V as alternate gene silencing products.

For independent verification of ALG10B (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-141010A, sc-141010B and sc-141010C.

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

ALG10B siRNA (m) is recommended for the inhibition of ALG10B 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ALG10B gene expression knockdown using RT-PCR Primer: ALG10B (m)-PR: sc-141010-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.