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SANTA CRUZ BIOTECHNOLOGY, INC.

Sar1B siRNA (m): sc-153222

BACKGROUND

There are a number of components involved in the secretory pathway of cells. Vesicular traffic within the early secretory pathway is mediated by COPI- and COPII-coated vesicles. The COPII vesicle coat protein promotes the formation of endoplasmic reticulum (ER) derived transport vesicles that carry secretory proteins to the Golgi complex. The SAR1 gene encodes two isoforms, Sar1a and Sar1B, in mammalian cells. These proteins are low-molecular-weight GTPases, which are essential for the formation of transport vesicles from the ER. Mutations in the SAR1 gene result in Anderson's disease (and/or chylomicron retention disease CMRD), a rare, autosomal recessive lipid malabsorption disorder characterized by chronic diarrhea, failure to thrive and hypocholesterolemia in childhood.

REFERENCES

1. Kuge, O., et al. 1994. Sar1 promotes vesicle budding from the endoplasmic reticulum but not Golgi compartments. J. Cell Biol. 125: 51-65.
2. Vahlensieck, Y., et al. 1995. Transcriptional studies on yeast SEC genes provide no evidence for regulation at the transcriptional level. Yeast 11: 901-911.
3. Salama, N.R., et al. 1997. Sec31 encodes an essential component of the COPII coat required for transport vesicle budding from the endoplasmic reticulum. Mol. Biol. Cell 8: 205-217.
4. Nickel, W., et al. 1998. Protein and lipid sorting between the endoplasmic reticulum and the Golgi complex. Semin. Cell Dev. Biol. 9: 493-501.
5. Saito, Y., et al. 1999. Identification of SEC12, SED4, truncated SEC16, and EKS1/HRD3 as multicopy suppressors of ts mutants of Sar1 GTPase. J. Biochem. 125: 130-137.
6. Shoulders, C.C., et al. 2004. The intracellular transport of chylomicrons requires the small GTPase, Sar1B. Curr. Opin. Lipidol. 15: 191-197.
7. Wang, X.M., et al. 2006. Sequence identification, tissue distribution, mapping and polymorphism of the porcine sar1b gene. Anim. Biotechnol. 17: 99-107.
8. Silvain, M., et al. 2008. Anderson's disease (chylomicron retention disease): a new mutation in the SARA2 gene associated with muscular and cardiac abnormalities. Clin. Genet. 74: 546-552.

CHROMOSOMAL LOCATION

Genetic locus: Sar1b (mouse) mapping to 11 B1.3.

PRODUCT

Sar1B 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Sar1B shRNA Plasmid (m): sc-153222-SH and Sar1B shRNA (m) Lentiviral Particles: sc-153222-V as alternate gene silencing products.

For independent verification of Sar1B (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-153222A, sc-153222B and sc-153222C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Sar1B siRNA (m) is recommended for the inhibition of Sar1B 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 µM in 66 µl. 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

Sar1B (AT1C7): sc-517425 is recommended as a control antibody for monitoring of Sar1B 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Sar1B gene expression knockdown using RT-PCR Primer: Sar1B (m)-PR: sc-153222-PR (20 µl). 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.