

SAC1 siRNA (h): sc-78323

BACKGROUND

The *Saccharomyces cerevisiae* SAC1 gene modulates yeast actin function and alleviates the essential requirement for phosphatidylinositol transfer protein (sec14p) activity in Golgi secretory function. The SAC1 gene product (Sac1p) is an integral membrane lipid phosphatase of the endoplasmic reticulum (ER) and the Golgi complex and contains a Sac phosphatase domain. Sac1p functions in a wide range of cellular processes including inositol metabolism, actin cytoskeletal organization, endoplasmic reticulum ATP transport, phosphatidylinositol-phosphatidylcholine transfer protein function and multiple-drug sensitivity. Sac1p is an important regulator of microsomal ATP transport, providing a link between inositol phospholipid signaling and ATP-dependent processes in the yeast ER. Defects in Sac1p relieves the requirement for Sec14p by altering phospholipid metabolism to expand the pool of diacylglycerol in the Golgi. Sac1p dysfunction exerts its pleiotropic effects on yeast Golgi function, the organization of the Actin cytoskeleton, and the cellular requirement for inositol, through altered metabolism of inositol glycerophospholipids. These effects suggest the secretory and cytoskeletal activities are coordinated to achieve proper spatial regulation of secretion in *S. cerevisiae*.

REFERENCES

1. Cleves, A.E., et al. 1989. Mutations in the SAC1 gene suppress defects in yeast Golgi and yeast Actin function. *J. Cell Biol.* 109: 2939-2950.
2. Whitters, E.A., et al. 1993. SAC1p is an integral membrane protein that influences the cellular requirement for phospholipid transfer protein function and inositol in yeast. *J. Cell Biol.* 122: 79-94.
3. Kearns, B.G., et al. 1997. Essential role for diacylglycerol in protein transport from the yeast Golgi complex. *Nature* 387: 101-105.
4. Kochendorfer, K.U., et al. 1999. Sac1p plays a crucial role in microsomal ATP transport, which is distinct from its function in Golgi phospholipid metabolism. *EMBO J.* 18: 1506-1515.
5. Hughes, W.E., et al. 2000. Sac phosphatase domain protein. *Biochem. J.* 350: 337-352.
6. Hughes, W.E., et al. 2000. SAC1 encodes a regulated lipid phosphoinositide phosphatase, defects in which can be suppressed by the homologous Inp52p and Inp53p phosphatases. *J. Biol. Chem.* 275: 801-808.

CHROMOSOMAL LOCATION

Genetic locus: SACM1L (human) mapping to 3p21.31.

PRODUCT

SAC1 siRNA (h) 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 SAC1 shRNA Plasmid (h): sc-78323-SH and SAC1 shRNA (h) Lentiviral Particles: sc-78323-V as alternate gene silencing products.

For independent verification of SAC1 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-78323A, sc-78323B and sc-78323C.

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

SAC1 siRNA (h) is recommended for the inhibition of SAC1 expression in human 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 SAC1 gene expression knockdown using RT-PCR Primer: SAC1 (h)-PR: sc-78323-PR (20 μ l, 477 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Dickson, E.J., et al. 2016. Dynamic formation of ER-PM junctions presents a lipid phosphatase to regulate phosphoinositides. *J. Cell Biol.* 213: 33-48.

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.