

SPPase2 siRNA (h): sc-63062

BACKGROUND

Sphingosine-1-phosphate (S1P) is a lipid that functions in a variety of intracellular and extracellular biological events. The intracellular activity of S1P is regulated by its state of phosphorylation and is therefore controlled by SPP kinases (SphKs) and S1P phosphatases (SPPases). The mammalian SPPases belong to the type 2 lipid phosphate phosphatase family of N-ethylmaleimide insensitive, magnesium-independent, multi-pass membrane proteins. Characteristic of their family, SPPases contain three conserved motifs that comprise the active site of the enzyme: the SXH motif, the KXXXXXXRP motif and the SRXXXXHXXD motif. SPPase2, also known as SGPP2 or SPP2, localizes to the membrane of the endoplasmic reticulum (ER) and is predominantly expressed in kidney and heart with lower levels found in colon, lung, brain and small intestine. SPPase2 specifically dephosphorylates S1P, dihydro-S1P and phyto-S1P. The overexpression of SPPases can lead to an elevation in the levels of ceramide and can induce apoptosis.

REFERENCES

1. Mandala, S.M. 2001. Sphingosine-1-phosphate phosphatases. *Prostaglandins Other Lipid Mediat.* 64: 143-156.
2. Cuvillier, O. 2002. Sphingosine in apoptosis signaling. *Biochim. Biophys. Acta* 1585: 153-162.
3. Le Stunff, H., Galve-Roperh, I., Peterson, C., Milstien, S. and Spiegel, S. 2002. Sphingosine-1-phosphate phosphohydrolase in regulation of sphingolipid metabolism and apoptosis. *J. Cell Biol.* 158: 1039-1049.
4. Ogawa, C., Kihara, A., Gokoh, M. and Igarashi, Y. 2003. Identification and characterization of a novel human sphingosine-1-phosphate phosphohydrolase, hSPP2. *J. Biol. Chem.* 278: 1268-1272.
5. Johnson, K.R., Johnson, K.Y., Becker, K.P., Bielawski, J., Mao, C. and Obeid, L.M. 2003. Role of human sphingosine-1-phosphate phosphatase 1 in the regulation of intra- and extracellular sphingosine-1-phosphate levels and cell viability. *J. Biol. Chem.* 278: 34541-34547.
6. Saba, J.D. and Hla, T. 2004. Point-counterpoint of sphingosine 1-phosphate metabolism. *Circ. Res.* 94: 724-734.
7. Pyne, S., Kong, K.C. and Darroch, P.I. 2004. Lysophosphatidic acid and sphingosine 1-phosphate biology: the role of lipid phosphate phosphatases. *Semin. Cell Dev. Biol.* 15: 491-501.
8. Le Stunff, H., Mikami, A., Giussani, P., Hobson, J.P., Jolly, P.S., Milstien, S. and Spiegel, S. 2004. Role of sphingosine-1-phosphate phosphatase 1 in epidermal growth factor-induced chemotaxis. *J. Biol. Chem.* 279: 34290-34297.
9. Wendler, C.C. and Rivkees, S.A. 2006. Sphingosine-1-phosphate inhibits cell migration and endothelial to mesenchymal cell transformation during cardiac development. *Dev. Biol.* 291: 264-277.

CHROMOSOMAL LOCATION

Genetic locus: SGPP2 (human) mapping to 2q36.1.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PRODUCT

SPPase2 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 SPPase2 shRNA Plasmid (h): sc-63062-SH and SPPase2 shRNA (h) Lentiviral Particles: sc-63062-V as alternate gene silencing products.

For independent verification of SPPase2 (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-63062A, sc-63062B and sc-63062C.

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

SPPase2 siRNA (h) is recommended for the inhibition of SPPase2 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 SPPase2 gene expression knockdown using RT-PCR Primer: SPPase2 (h)-PR: sc-63062-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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