

SPPase1 siRNA (h): sc-63060

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. SPPase1, also known as SGPP1 or SPP1, localizes to the membrane of the endoplasmic reticulum (ER) and is ubiquitously expressed with highest levels found in kidney and placenta. SPPase1 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* 64: 143-156.
2. Cuvillier, O. 2002. Sphingosine in apoptosis signaling. *Biochim. Biophys. Acta* 1585: 153-162.
3. Le Stunff, H., et al. 2002. Sphingosine-1-phosphate phosphohydrolase in regulation of sphingolipid metabolism and apoptosis. *J. Cell Biol.* 158: 1039-1049.
4. Ogawa, C., et al. 2003. Identification and characterization of a novel human sphingosine-1-phosphate phosphohydrolase, hSPP2. *J. Biol. Chem.* 278: 1268-1272.
5. Johnson, K.R., et al. 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.

CHROMOSOMAL LOCATION

Genetic locus: SGPP1 (human) mapping to 14q23.2.

PRODUCT

SPPase1 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 SPPase1 shRNA Plasmid (h): sc-63060-SH and SPPase1 shRNA (h) Lentiviral Particles: sc-63060-V as alternate gene silencing products.

For independent verification of SPPase1 (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-63060A, sc-63060B and sc-63060C.

RESEARCH USE

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

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

SPPase1 siRNA (h) is recommended for the inhibition of SPPase1 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 SPPase1 gene expression knockdown using RT-PCR Primer: SPPase1 (h)-PR: sc-63060-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Huang, X., et al. 2013. miRNA-95 mediates radioresistance in tumors by targeting the sphingolipid phosphatase SGPP1. *Cancer Res.* 73: 6972-6986.

PROTOCOLS

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