

N-SMase3 siRNA (h): sc-95034

BACKGROUND

Sphingomyelin and its metabolic products are well known to have second messenger functions in a variety of cellular signaling pathways. At the epicenter of the sphingomyelin cell signaling pathway is a family of phospholipases called sphingomyelinases. These enzymes cleave sphingomyelin to produce ceramide and phosphocholine, two proteins that mediate cell growth arrest and apoptosis. N-SMase3 (neutral sphingomyelinase 3), also known as NSMASE3, nSMase-3 or SMPD4, is an 827 amino acid single-pass membrane protein that localizes to the endoplasmic reticulum and Golgi apparatus. Widely expressed in most tissues, with highest levels in heart and skeletal muscle, N-SMase3 is a member of the magnesium-dependent phosphohydrolase protein family. Enhanced in the presence of phosphatidylserine and tumor necrosis factor (TNF) and inhibited by scyphostatin, N-SMase3 exhibits optimal activity at pH 7. N-SMase3 is considered to play an important role in tumorigenesis and cellular stress response.

REFERENCES

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6. Marchesini, N., et al. 2003. Biochemical properties of mammalian neutral sphingomyelinase 2 and its role in sphingolipid metabolism. J. Biol. Chem. 278: 13775-13783.
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CHROMOSOMAL LOCATION

Genetic locus: SMPD4 (human) mapping to 2q21.1.

PROTOCOLS

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

PRODUCT

N-SMase3 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 N-SMase3 shRNA Plasmid (h): sc-95034-SH and N-SMase3 shRNA (h) Lentiviral Particles: sc-95034-V as alternate gene silencing products.

For independent verification of N-SMase3 (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-95034A, sc-95034B and sc-95034C.

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

N-SMase3 siRNA (h) is recommended for the inhibition of N-SMase3 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 N-SMase3 gene expression knockdown using RT-PCR Primer: N-SMase3 (h)-PR: sc-95034-PR (20 μ l, 561 bp). 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.