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# ASM siRNA (h): sc-41650

## BACKGROUND

Acid sphingomyelinase (ASM) is a lysosomal protein that hydrolyzes sphingomyelin to ceramide and phosphocholine. The ASM gene encodes three proteins, ASM-1, ASM-2 and ASM-3, of which ASM-1 is the only ASM gene product that is a catalytically active enzyme. Deficiency of ASM is associated with type A and type B Niemann-Pick disease. Type A is a fatal neurodegenerative disorder seen in infancy and resulting in death by age three, whereas type B is a non-neuropathic disease that has a later onset. During monocytic cell differentiation, the expression of ASM is upregulated by the combined actions of AP-2 and Sp1 transcription factors.

## REFERENCES

1. Quintern, L.E., et al. 1987. Acid sphingomyelinase from human urine: purification and characterization. *Biochim. Biophys. Acta* 922: 323-336.
2. Schuchman, E.H., et al. 1991. Human acid sphingomyelinase. Isolation, nucleotide sequence and expression of the full-length and alternatively spliced cDNAs. *J. Biol. Chem.* 266: 8531-8539.
3. Levran, O., et al. 1991. Niemann-Pick disease: a frequent missense mutation in the acid sphingomyelinase gene of Ashkenazi Jewish type A and B patients. *Proc. Natl. Acad. Sci. USA* 88: 3748-3752.
4. Takahashi, T., et al. 1992. Identification and expression of five mutations in the human acid sphingomyelinase gene causing types A and B Niemann-Pick disease. Molecular evidence for genetic heterogeneity in the neuropathic and non-neuropathic forms. *J. Biol. Chem.* 267: 12552-12558.

## CHROMOSOMAL LOCATION

Genetic locus: SMPD1 (human) mapping to 11p15.4.

## PRODUCT

ASM siRNA (h) is a target-specific 19-25 nt siRNA designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ASM shRNA Plasmid (h): sc-41650-SH and ASM shRNA (h) Lentiviral Particles: sc-41650-V as alternate gene silencing products.

## STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at  $-20^{\circ}$  C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at  $-20^{\circ}$  C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

ASM siRNA (h) is recommended for the inhibition of ASM 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  $\mu$ M in 66  $\mu$ 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.

## GENE EXPRESSION MONITORING

ASM (4H2): sc-293189 is recommended as a control antibody for monitoring of ASM 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-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>TM</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ASM gene expression knockdown using RT-PCR Primer: ASM (h)-PR: sc-41650-PR (20  $\mu$ l, 428 bp). Annealing temperature for the primers should be 55-60 $^{\circ}$  C and the extension temperature should be 68-72 $^{\circ}$  C.

## SELECT PRODUCT CITATIONS

1. Garnett, T.O., et al. 2007. Bid is cleaved upstream of caspase-8 activation during TRAIL-mediated apoptosis in human osteosarcoma cells. *Apoptosis* 12: 1299-1315.
2. Bennaceur, K., et al. 2009. Different mechanisms are involved in apoptosis induced by melanoma gangliosides on human monocyte-derived dendritic cells. *Glycobiology* 19: 576-582.
3. Bao, J.X., et al. 2012. Lysosome-membrane fusion mediated superoxide production in hyperglycaemia-induced endothelial dysfunction. *PLoS ONE* 7: e30387.
4. Getz, T., et al. 2016. Quantum dot-mediated delivery of siRNA to inhibit sphingomyelinase activities in brain-derived cells. *J. Neurochem.* 139: 872-885.

## RESEARCH USE

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