SANTA CRUZ BIOTECHNOLOGY, INC.

HEXA siRNA (h): sc-60783

BACKGROUND

Hexosaminidase A (HEXA), also designated β-Hexosaminidase A, is a trimer composed of one α chain, one β-A chain and one β-B chain and is found in the lysosomes of cells. HEXA, along with the cofactor CM^2 activator protein, catalyzes the degradation of GM2 ganglioside and other molecules containing terminal N-acetyl hexosamines in the brain and other tissues. A mutation in the α subunit of hexosaminidase is the cause of Tay-Sachs disease (TSD), also known as GM2-gangliosidosis type I. TSD is a fatal autosomal recessive lysosomal storage disease of the central nervous system (CNS) caused by insufficient activity of the HEXA enzyme that results in a failure to process GM2 gangliosides. The accumulation of GM2 ganglioside in the absence of HEXA activity causes progressive destruction of the CNS.

REFERENCES

1. Triggs-Raine, B.L., et al. 1991. Sequence of DNA flanking the exons of the HEXA gene, and identification of mutations in Tay-Sachs disease. Am. J. Hum. Genet. 49: 1041-1054.
2. Yamanaka, S., et al. 1994. Targeted disruption of the Hexa gene results in mice with biochemical and pathologic features of Tay-Sachs disease. Proc. National Acad. .Sci. USA 91: 9975-9979.
3. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 606869. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Yamamoto, N. and Urade, M. 2005. Pathogenic significance of α-N-acetylgalactosaminidase activity found in the hemagglutinin of influenza virus. Microbes Infect. 7: 674-681.
5. Sanon, A., et al. 2005. N-acetyl-β-D-hexosaminidase from *Trichomonas vaginalis:* sub and activity of inhibitors. Biomed. Pharmacother. 59: 245-248.
6. Casal, J.A., et al. 2005. β-hexosaminidase isoenzyme profile polymorphonuclear and unfractionated total leukocytes. Clin. Biochem. 38: 938-942.

CHROMOSOMAL LOCATION

Genetic locus: HEXA (human) mapping to 15q23.

PRODUCT

HEXA 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HEXA shRNA Plasmid (h): sc-60783-SH and HEXA shRNA (h) Lentiviral Particles: sc-60783-V as alternate gene silencing products.

For independent verification of HEXA (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-60783A, sc-60783B and sc-60783C.

PROTOCOLS

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

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

HEXA siRNA (h) is recommended for the inhibition of HEXA 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 µM in 66 µl. 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

HEXA (D-2): sc-376777 is recommended as a control antibody for monitoring of HEXA 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-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HEXA gene expression knockdown using RT-PCR Primer: HEXA (h)-PR: sc-60783-PR (20 µl 445 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.