

HEXB siRNA (m): sc-60786

BACKGROUND

Hexosaminidase B (HEXB), also designated β -hexosaminidase B, is a tetramer of two β -A and two β -B chains and is found in the lysosomes of cells. Sandhoff disease (SD), also known as GM2-gangliosidosis type II, is caused by mutations in the HEXB gene that affect the b subunit. These mutations disrupt the activity of HEXB and HEXA, which prevents the breakdown of GM2 ganglioside, a fatty material found in the brain, thereby rendering both the HEXA and HEXB enzymes deficient. SD is a rare autosomal recessive disorder characterized by an accumulation of GM2 ganglioside, which causes progressive destruction of the central nervous system. Sandhoff disease is similar to Tay-Sachs disease, which is caused by mutations in the HEXA gene, although SD is more severe.

REFERENCES

1. Beutler, E., et al. 1975. Hexosaminidase isozyme in type O GM2 gangliosidosis (Sandhoff-Jatzkewitz disease). *Am. J. Hum. Genet.* 27: 628-638.
2. O'Dowd, B.F., et al. 1985. Isolation of cDNA clones coding for the β subunit of human β -hexosaminidase. *Proc. Natl. Acad. Sci. USA* 82: 1184-1188.
3. Bikker, H., et al. 1989. Demonstration of a Sandhoff disease-associated autosomal 50-kb deletion by field inversion gel electrophoresis. *Hum. Genet.* 81: 287-288.
4. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 606873. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Yamamoto, N. and Urade, M. 2005. Pathogenic significance of α -N-acetyl-galactosaminidase activity found in the hemagglutinin of influenza virus. *Microbes Infect.* 7: 674-681.
6. Sanon, A., et al. 2005. N-acetyl- β -D-hexosaminidase from *Trichomonas vaginalis*: sub and activity of inhibitors. *Biomed. Pharmacother.* 59: 245-248.
7. Casal, J.A., et al. 2005. β -hexosaminidase isoenzyme profile polymorphonuclear and unfractionated total leukocytes. *Clin. Biochem.* 38: 938-942.

CHROMOSOMAL LOCATION

Genetic locus: Hexb (mouse) mapping to 13 D1.

PRODUCT

HEXB siRNA (m) 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 HEXB shRNA Plasmid (m): sc-60786-SH and HEXB shRNA (m) Lentiviral Particles: sc-60786-V as alternate gene silencing products.

For independent verification of HEXB (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-60786A, sc-60786B and sc-60786C.

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

HEXB siRNA (m) is recommended for the inhibition of HEXB expression in mouse 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.

GENE EXPRESSION MONITORING

HEXB B chain (D-9): sc-376781 is recommended as a control antibody for monitoring of HEXB 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 κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 HEXB gene expression knockdown using RT-PCR Primer: HEXB (m)-PR: sc-60786-PR (20 μ l). 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.

PROTOCOLS

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