

iGb3 siRNA (m): sc-146179

BACKGROUND

iGb3 (isogloboside 3), also known as isoglobotrihexosylceramide, A3GALT2 (α 1,3-galactosyltransferase 2), A3GALT2P or IGBS3S (isoglobotriaosylceramide synthase), is a 370 amino acid protein that belongs to the glycosyltransferase 6 family. Involved in natural killer T cell development, iGb3 also synthesizes the galactose- α (1,3)-galactose group on α -galactose. Localizing to Golgi apparatus, iGb3 glycosylates lipids and binds manganese as a cofactor. The gene encoding iGb3 maps to human chromosome 1p35.1, which spans 260 million base pairs and contains over 3,000 genes. Human chromosome 1 comprises nearly 8% of the human genome and houses a large number of disease-associated genes, including those that are involved in familial adenomatous polyposis, Stickler syndrome, Parkinson's disease, Gaucher disease, schizophrenia and Usher syndrome.

REFERENCES

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2. Lau, E.K., et al. 1999. Two novel polymorphic sequences in the glucocerebrosidase gene region enhance mutational screening and founder effect studies of patients with Gaucher disease. *Hum. Genet.* 104: 293-300.
3. Plasilova, M., et al. 2004. Exclusion of an extracolonic disease modifier locus on chromosome 1p33-36 in a large Swiss familial adenomatous polyposis kindred. *Eur. J. Hum. Genet.* 12: 365-371.
4. Oliveira, S.A., et al. 2005. Identification of risk and age-at-onset genes on chromosome 1p in Parkinson disease. *Am. J. Hum. Genet.* 77: 252-264.
5. Porubsky, S., et al. 2007. Normal development and function of invariant natural killer T cells in mice with isoglobotrihexosylceramide (iGb3) deficiency. *Proc. Natl. Acad. Sci. USA* 104: 5977-5982.
6. Christiansen, D., et al. 2008. Humans lack iGb3 due to the absence of functional iGb3-synthase: implications for NKT cell development and transplantation. *PLoS Biol.* 6: e172.
7. Yurov, Y.B., et al. 2008. The schizophrenia brain exhibits low-level aneuploidy involving chromosome 1. *Schizophr. Res.* 98: 139-147.
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CHROMOSOMAL LOCATION

Genetic locus: A3galt2 (mouse) mapping to 4 D2.2.

PRODUCT

iGb3 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 iGb3 shRNA Plasmid (m): sc-146179-SH and iGb3 shRNA (m) Lentiviral Particles: sc-146179-V as alternate gene silencing products.

For independent verification of iGb3 (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-146179A, sc-146179B and sc-146179C.

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

iGb3 siRNA (m) is recommended for the inhibition of iGb3 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

iGb3 (H-7): sc-514782 is recommended as a control antibody for monitoring of iGb3 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 iGb3 gene expression knockdown using RT-PCR Primer: iGb3 (m)-PR: sc-146179-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.