

NEGR1 siRNA (m): sc-149901

BACKGROUND

NEGR1 (neuronal growth regulator 1), also known as neurotractin, IGLON4 (IgLN family member 4), Ntra, DMML2433 or KILON, is a 354 amino acid protein belonging to the IgLN family and immunoglobulin superfamily. NEGR1 may play a role in cell adhesion and regenerative axon sprouting in the mammalian brain and is highly expressed in adult hippocampus, cerebellum and brainstem, with much lower levels found in cerebellum. Localizing to the cell membrane at the glycosylphosphatidylinositol anchor (GPI) anchor, NEGR1 contains three Ig-like C2-type (immunoglobulin-like) domains. NEGR1 is encoded by a gene that maps to human chromosome 1p31.1, and is one of several loci associated with body mass index (BMI), possibly contributing to the development of obesity.

REFERENCES

- Funatsu, N., et al. 1999. Characterization of a novel rat brain glycosylphosphatidylinositol-anchored protein (Kilon), a member of the IgLN cell adhesion molecule family. *J. Biol. Chem.* 274: 8224-8230.
- Miyata, S., et al. 2003. Biochemical and ultrastructural analyses of IgLN cell adhesion molecules, Kilon and OBCAM in the rat brain. *Neuroscience* 117: 645-658.
- Schäfer, M., et al. 2005. Neurotractin/kilon promotes neurite outgrowth and is expressed on reactive astrocytes after entorhinal cortex lesion. *Mol. Cell. Neurosci.* 29: 580-590.
- Bauer, F., et al. 2009. Obesity genes identified in genome-wide association studies are associated with adiposity measures and potentially with nutrient-specific food preference. *Am. J. Clin. Nutr.* 90: 951-959.
- Thorleifsson, G., et al. 2009. Genome-wide association yields new sequence variants at seven loci that associate with measures of obesity. *Nat. Genet.* 41: 18-24.
- Willer, C.J., et al. 2009. Six new loci associated with body mass index highlight a neuronal influence on body weight regulation. *Nat. Genet.* 41: 25-34.
- Online Mendelian Inheritance in Man, OMIM[™]. 2009. Johns Hopkins University, Baltimore, MD. MIM Number: 613173. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>

CHROMOSOMAL LOCATION

Genetic locus: Negr1 (mouse) mapping to 3 H4.

PRODUCT

NEGR1 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 NEGR1 shRNA Plasmid (m): sc-149901-SH and NEGR1 shRNA (m) Lentiviral Particles: sc-149901-V as alternate gene silencing products.

For independent verification of NEGR1 (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-149901A, sc-149901B and sc-149901C.

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

NEGR1 siRNA (m) is recommended for the inhibition of NEGR1 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

NEGR1 (H-12): sc-393293 is recommended as a control antibody for monitoring of NEGR1 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 NEGR1 gene expression knockdown using RT-PCR Primer: NEGR1 (m)-PR: sc-149901-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.