SANTA CRUZ BIOTECHNOLOGY, INC.

Nir2 siRNA (m): sc-40854

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The Power to Question

BACKGROUND

The Nirs (Nir1, Nir2, and Nir3), human homologues of *Drosophila* retinal degeneration B (RdgB), have been considered candidate genes for human inherited retinal degeneration diseases. The three Nir proteins are highly expressed in the developing retina, each exhibiting a distinct distribution profile. Immunolocalization studies revealed that Nir2 is mainly localized in the Golgi apparatus in interphase cells, but it is recruited to the cleavage furrow and the midbody during cytokinesis. Additionally, Nir2, like RdgB, contains an amino-terminal phosphatidylinositol-transfer protein (PITP)-like domain and is essential for cytokinesis. In contrast to related PITP proteins, the RdgB proteins, which include Nir2 and Nir3, contain an amino-terminal PITP-like domain, an acidic, calcium-binding domain, six putative transmembrane domains and a conserved carboxyl-terminal domain. It has been suggested that Nir and RdgB proteins represent a new family of evolutionarily conserved PYK2-binding proteins that play a role in the control of calcium and phosphoinositide metabolism downstream of G protein-coupled receptors.

REFERENCES

1. Fullwood, Y., dos Santos, M. and Hsuan, J.J. 1999. Cloning and characterization of a novel human phosphatidylinositol transfer protein, RdgB β. J. Biol. Chem. 274: 31553-31558.
2. Tian, D. and Lev, S. 2002. Cellular and developmental distribution of human homologues of the *Drosophilia* RdgB protein in the rat retina. Invest. Ophthalmol. Vis. Sci. 43: 1946-1953.
3. Tian, D., Litvak, V., Toledo-Rodriguez, M., Carmon, S. and Lev, S 2002. Nir2, a novel regulator of cell morphogenesis. Mol. Cell. Biol. 22: 2650-2662.
4. Litvak, V., Tian, D., Carmon, S. and Lev, S. 2002. Nir2, a human homolog of *Drosophila* melanogaster retinal degeneration B protein, is essential for cytokinesis. Mol. Cell. Biol. 22: 5064-5075.
5. Litvak, V., Shaul, Y.D., Shulewitz, M., Amarilio, R., Carmon, S. and Lev, S. 2002. Targeting of Nir2 to lipid droplets is regulated by a specific threonine residue within its PI-transfer domain. Curr. Biol. 12: 1513-1518.

CHROMOSOMAL LOCATION

Genetic locus: Pitpnm1 (mouse) mapping to 19 A.

PRODUCT

Nir2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Nir2 shRNA Plasmid (m): sc-40854-SH and Nir2 shRNA (m) Lentiviral Particles: sc-40854-V as alternate gene silencing products.

For independent verification of Nir2 (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-40854A, sc-40854B and sc-40854C.

RESEARCH USE

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

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

Nir2 siRNA (m) is recommended for the inhibition of Nir2 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 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

Nir2 (8): sc-136140 is recommended as a control antibody for monitoring of Nir2 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 Nir2 gene expression knockdown using RT-PCR Primer: Nir2 (m)-PR: sc-40854-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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