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NCDN siRNA (m): sc-149854

BACKGROUND

NCDN (neurochondrin) is a 729 amino acid leucine-rich cytoplasmic protein belonging to the neurochondrin family that is involved in nervous system signal transduction and required for spatial learning. Known to act as a negative regulator of CaMKII (Ca²⁺-calmodulin-dependent protein kinase 2) phosphorylation, NCDN may also associate with MCH-1R (melanin-concentrating hormone receptor 1) to modulate its function, and is suggested to play a role in bone metabolism, neurite outgrowth and chondrocyte differentiation. Due to alternative splicing events, three NCDN isoforms are known to exist which are highly expressed in adult brain and spinal cord, and found at lower levels in fetal brain, ovary and testis. Localizing to somatic regions of neurons, NCDN is encoded by a gene that maps to human chromosome 1p34.3 and mouse chromosome 4 D2.2.

REFERENCES

1. Nagase, T., et al. 1998. Prediction of the coding sequences of unidentified human genes. IX. The complete sequences of 100 new cDNA clones from brain which can code for large proteins *in vitro*. DNA Res. 5: 31-39.
2. Mochizuki, R., et al. 1999. Molecular cloning and expression of human neurochondrin-1 and -2. Biochim. Biophys. Acta 1446: 397-402.
3. Mochizuki, R., et al. 2000. Corrigendum to "Molecular cloning and expression of human neurochondrin-1 and -2". Biochim. Biophys. Acta 1490: 367-368.
4. Mochizuki, R., et al. 2003. Targeted disruption of the neurochondrin/norbin gene results in embryonic lethality. Biochem. Biophys. Res. Commun. 310: 1219-1226.
5. Dateki, M., et al. 2005. Neurochondrin negatively regulates CaMKII phosphorylation, and nervous system-specific gene disruption results in epileptic seizure. J. Biol. Chem. 280: 20503-20508.

CHROMOSOMAL LOCATION

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

PRODUCT

NCDN 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 NCDN shRNA Plasmid (m): sc-149854-SH and NCDN shRNA (m) Lentiviral Particles: sc-149854-V as alternate gene silencing products.

For independent verification of NCDN (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-149854A, sc-149854B and sc-149854C.

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 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

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

NCDN (B-3): sc-398686 is recommended as a control antibody for monitoring of NCDN 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 NCDN gene expression knockdown using RT-PCR Primer: NCDN (m)-PR: sc-149854-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.