

Ndn siRNA (h): sc-37318

BACKGROUND

Prader-Willi syndrome (PWS) is a neurogenetic disorder resulting from the loss of paternal expression of gene(s) localized in the 15q11.2 region. Clinical manifestations of this disease include feeding problems in infancy, temper outbursts, perseveration, obsessive-compulsive symptoms and sleep disturbances. Necdin (Ndn) protein is generated from an intronless gene that is located in the Prader-Willi syndrome deletion region. Studies in mouse suggest that the protein encoded by this gene may suppress growth in postmitotic neurons. Ndn expression in brain is restricted to post-mitotic neurons and parental alleles display a differential methylation profile in the coding region. Reduced expression of Ndn is responsible for at least a subset of the clinical manifestations of PWS, including skin picking and improved spatial memory.

REFERENCES

1. Watrin, F., et al. 1997. The mouse Necdin gene is expressed from the paternal allele only and lies in the 7C region of the mouse chromosome 7, a region of conserved synteny to the human Prader-Willi syndrome region. *Eur. J. Hum. Genet.* 5: 324-332.
2. Jay, P., et al. 1997. The human Necdin gene, NDN, is maternally imprinted and located in the Prader-Willi syndrome chromosomal region. *Nat. Genet.* 17: 357-361.
3. Muscatelli, F., et al. 2000. Disruption of the mouse Necdin gene results in hypothalamic and behavioral alterations reminiscent of the human Prader-Willi syndrome. *Hum. Mol. Genet.* 9: 3101-3110.

CHROMOSOMAL LOCATION

Genetic locus: NDN (human) mapping to 15q11.2.

PRODUCT

Ndn siRNA (h) 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 Ndn shRNA Plasmid (h): sc-37318-SH and Ndn shRNA (h) Lentiviral Particles: sc-37318-V as alternate gene silencing products.

For independent verification of Ndn (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-37318A, sc-37318B and sc-37318C.

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

Ndn siRNA (h) is recommended for the inhibition of Ndn expression in human 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

Ndn (36-V): sc-101224 is recommended as a control antibody for monitoring of Ndn 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 Ndn gene expression knockdown using RT-PCR Primer: Ndn (h)-PR: sc-37318-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ingraham, C.A., et al. 2011. Necdin and neurotrophin receptors: interactors of relevance for neuronal resistance to oxidant stress. *Pediatr. Res.* 69: 279-284.

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.