

Neuro D2 siRNA (h): sc-94081

BACKGROUND

Members of the myogenic determination family are basic helix-loop-helix (bHLH) proteins that can be separated into two classes, both of which work together to activate DNA transcription. Class A proteins include the ubiquitously expressed E-box binding factors, namely E2A, ITF-2 and HEB, while class B proteins, such as MyoD, myogenin and Neuro D (β 2), are transiently expressed and exhibit a much more limited tissue distribution. Working in opposition to these positively acting factors are a specialized group of basic helix-loop-helix (bHLH) transcription factors that function as dominant negative regulators and are involved in cell lineage determination and differentiation. Neuro D2 (neurogenic differentiation 2), also known as NDRF, NEUROD2 or bHLHA1, is a 382 amino acid nuclear protein that contains one bHLH domain and functions to induce neurogenic differentiation, playing an important role in the maintenance and determination of cell fate.

REFERENCES

1. McCormick, M.B., et al. 1996. Neuro D2 and Neuro D3: distinct expression patterns and transcriptional activation potentials within the Neuro D gene family. *Mol. Cell. Biol.* 16: 5792-5800.
2. Tamimi, R.M., et al. 1997. Neuro D2 and Neuro D3 genes map to human chromosomes 17q12 and 5q23-q31 and mouse chromosomes 11 and 13, respectively. *Genomics* 40: 355-357.
3. Online Mendelian Inheritance in Man, OMIM[™]. 1997. Johns Hopkins University, Baltimore, MD. MIM Number: 601725. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Kume, H., et al. 1998. Phosphorylation and spatiotemporal distribution of KW8 (NDRF/Neuro D2), a Neuro D family basic helix-loop-helix protein. *Brain Res. Mol. Brain Res.* 60: 107-114.
5. Shibata, H., et al. 1999. Interaction of PKN with a Neuron-specific basic helix-loop-helix transcription factor, NDRF/Neuro D2. *Brain Res. Mol. Brain Res.* 74: 126-134.

CHROMOSOMAL LOCATION

Genetic locus: NEUROD2 (human) mapping to 17q12.

PRODUCT

Neuro D2 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 Neuro D2 shRNA Plasmid (h): sc-94081-SH and Neuro D2 shRNA (h) Lentiviral Particles: sc-94081-V as alternate gene silencing products.

For independent verification of Neuro D2 (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-94081A, sc-94081B and sc-94081C.

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

Neuro D2 siRNA (h) is recommended for the inhibition of Neuro D2 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

Neuro D2 (G-10): sc-365896 is recommended as a control antibody for monitoring of Neuro D2 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 Neuro D2 gene expression knockdown using RT-PCR Primer: Neuro D2 (h)-PR: sc-94081-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.