

atrophin-1 siRNA (h): sc-29765

BACKGROUND

Dentatorubral-pallidoluysian atrophy protein, also designated atrophin-1, interacts with several other proteins, including RERE, BAIAP2 and WWP1-3. It is highly expressed in ovary, testis, brain and prostate, but can also be detected in thymus, liver and leukocytes. Defects in ATN1, the gene encoding for the atrophin protein, can cause dentatorubral-pallidoluysian atrophy (DRPLA) or Haw River syndrome (HRS). Both disorders are dominant neurodegenerative disorders caused by an increase in the number of polyglutamine (Gln) repeats in the ATN1 gene (7-23 repeats in the normal population, 49-75 in patients affected by DRPLA or HRS). More repeats corresponds to earlier onset and more severe clinical manifestations of the diseases. DRPLA is characterized by a loss of neurons in the dentate nucleus, rubrum, globus pallidus and Luys' body, often resulting in dementia, epilepsy and cerebellar ataxia. HRS is characterized by the degeneration of multiple systems, resembling that of DRPLA or Huntington's disease.

REFERENCES

1. Nagafuchi, S., et al. 1994. Structure and expression of the gene responsible for the triplet repeat disorder, dentatorubral and pallidoluysian atrophy (DRPLA). *Nat. Genet.* 8: 177-182.
2. Yazawa, I., et al. 1995. Abnormal gene product identified in hereditary dentatorubral-pallidoluysian atrophy (DRPLA) brain. *Nat. Genet.* 10: 99-103.
3. Miyashita, T., et al. 1997. Dentatorubral pallidoluysian atrophy (DRPLA) protein is cleaved by caspase-3 during apoptosis. *J. Biol. Chem.* 272: 29238-29242.
4. Wood, J.D., et al. 1998. Atrophin-1, the DRPLA gene product, interacts with two families of WW domain-containing proteins. *Mol. Cell. Neurosci.* 11: 149-160.
5. Kanazawa, I. 1998. Dentatorubral-pallidoluysian atrophy or Naito-Oyanagi disease. *Neurogenetics* 2: 1-17.
6. Schilling, G., et al. 1999. Nuclear accumulation of truncated atrophin-1 fragments in a transgenic mouse model of DRPLA. *Neuron* 24: 275-286.

CHROMOSOMAL LOCATION

Genetic locus: ATN1 (human) mapping to 12p13.31.

PRODUCT

atrophin-1 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 atrophin-1 shRNA Plasmid (h): sc-29765-SH and atrophin-1 shRNA (h) Lentiviral Particles: sc-29765-V as alternate gene silencing products.

For independent verification of atrophin-1 (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-29765A, sc-29765B and sc-29765C.

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 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

atrophin-1 siRNA (h) is recommended for the inhibition of atrophin-1 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

atrophin-1 (C1): sc-517594 is recommended as a control antibody for monitoring of atrophin-1 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 atrophin-1 gene expression knockdown using RT-PCR Primer: atrophin-1 (h)-PR: sc-29765-PR (20 μ l). 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.