

TorsinA siRNA (h): sc-42303

BACKGROUND

A mutation of the DYT1 gene, which codes for TorsinA, has been identified as the cause of one form of autosomal dominantly inherited dystonia. Early-onset torsion dystonia is a movement disorder, characterized by twisting muscle contractures, that begins in childhood. Symptoms are believed to result from altered neuronal communication in the basal ganglia. TorsinA comprises 332 amino acids and is widely expressed throughout the mouse central nervous system; it is detected in the majority of neurons in nearly all regions. The proteins display cytoplasmic distribution, although in some types of neurons localization is perinuclear. TorsinA often performs chaperone-like functions that assist in the assembly, operation, or dis-assembly of protein complexes. The gene which encodes TorsinA has high homology to three additional mammalian genes and a nematode gene and distal similarity to the family of heat-shock proteins and the Clp protease family. The gene which encodes TorsinA maps to human chromosome 9q34.11.

REFERENCES

1. Ozelius, L.J., et al. 1997. The early-onset torsion dystonia gene (DYT1) encodes an ATP-binding protein. *Nat. Genet.* 17: 40-48.
2. Neuwald, A.F., et al. 1999. AAA⁺: a class of chaperone-like ATPases associated with the assembly, operation, and disassembly of protein complexes. *Genome Res.* 9: 27-43.
3. Konakova, M. and Pulst, S.M. 2001. Immunocytochemical characterization of Torsin proteins in mouse brain. *Brain Res.* 922: 1-8.
4. Walker, R.H., et al. 2002. TorsinA immunoreactivity in brains of patients with DYT1 and non-DYT1 dystonia. *Neurology* 58: 120-124.
5. LocusLink Report (LocusID: 605204).
<http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: TOR1A (human) mapping to 9q34.11.

PRODUCT

TorsinA 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 TorsinA shRNA Plasmid (h): sc-42303-SH and TorsinA shRNA (h) Lentiviral Particles: sc-42303-V as alternate gene silencing products.

For independent verification of TorsinA (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-42303A, sc-42303B and sc-42303C.

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.

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

TorsinA siRNA (h) is recommended for the inhibition of TorsinA 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

TorsinA (D-7): sc-373915 is recommended as a control antibody for monitoring of TorsinA 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 TorsinA gene expression knockdown using RT-PCR Primer: TorsinA (h)-PR: sc-42303-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.