SANTA CRUZ BIOTECHNOLOGY, INC.

Ataxin-2 siRNA (h): sc-40356

BACKGROUND

Autosomal dominant cerebellar ataxias are a group of neurodegenerative disorders caused by unstable CAG repeat expansions encoding polyglutamine tracts. Proteins with long polyglutamine tracts have an increased tendency to aggregate, often forming ubiquitinated intranuclear inclusion bodies. Ataxin-2, the gene product of the human spinocerebellar ataxia type 2 (SCA2/ATXN2) gene, is a basic protein with two domains (Sm1 and Sm2) implicated in RNA splicing and protein interaction. Ataxin-2 interacts with a putative RNA-binding protein Ataxin-2-binding-protein 1 (A2BP1), which is expressed in muscle and brain. Ataxin-2 is ubiquitously expressed with highest levels in the cytoplasm of Purkinje cells. Both A2BP1 and Ataxin-2 are localized to the *trans*-Golgi network. Mice expressing Ataxin-2 with polyglutamine show progressive functional deficits accompanied by loss of Purkinje cell dendritic arbor and eventually loss of Purkinje cells. In conclusion, expansion of Ataxin-2 results in spinocerebellar ataxia type 2, which affects the cerebellum and other areas of the central nervous system.

REFERENCES

1. Gispert, S., et. al. 1993. Chromosomal assignment of the second locus for autosomal dominant cerebellar ataxia (SCA2) to chromosome 12q23-24.1 Nat. Genet. 4: 295-299.
2. Sanpei, K., et. al. 1996. Identification of the spinocerebellar ataxia type 2 gene using a direct identification of repeat expansion and cloning technique, DIRECT. Nat. Genet. 14: 277-284.
3. Pujana, M.A., et al. 1999. Spinocerebellar ataxias in Spanish patients: genetic analysis of familial and sporadic cases. The Ataxia Study Group. Hum. Genet. 104: 516-522.
4. Huynh, D.P., et al. 2000. Nuclear localization or inclusion body formation of Ataxin-2 are not necessary for SCA2 pathogenesis in mouse or human. Nat. Genet. 26: 44-50.
5. Huynh, D.P., et al. 2000. Expression of Ataxin-2 in brains from normal individuals and patients with Alzheimer's disease and spinocerebellar Ataxia-2. Ann. Neurol. 45: 232-241.
6. Shibata, H., et al. 2000. A novel protein with RNA-binding motifs interacts with Ataxin-2. Hum. Mol. Genet. 9: 1303-1313.

CHROMOSOMAL LOCATION

Genetic locus: ATXN2 (human) mapping to 12q24.12.

PRODUCT

Ataxin-2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Ataxin-2 shRNA Plasmid (h): sc-40356-SH and Ataxin-2 shRNA (h) Lentiviral Particles: sc-40356-V as alternate gene silencing products.

For independent verification of Ataxin-2 (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-40356A, sc-40356B and sc-40356C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Ataxin-2 siRNA (h) is recommended for the inhibition of Ataxin-2 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 µM in 66 µl. 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

Ataxin-2 (A-6): sc-515602 is recommended as a control antibody for monitoring of Ataxin-2 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Ataxin-2 gene expression knockdown using RT-PCR Primer: Ataxin-2 (h)-PR: sc-40356-PR (20 µl, 422 bp). 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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.