SANTA CRUZ BIOTECHNOLOGY, INC.

SMN siRNA (h): sc-36510

BACKGROUND

Spinal muscular atrophy (SMA) is an autosomal recessive neurodegenerative disease characterized by loss of motor neurons in the spinal cord. SMA is caused by deletion or loss-of-function mutations of SMN (survival of motor neuron) gene. SMN, also known as Gemin1, SMN1, SMNT and BCD541, exists as four isoforms produced by alternative splicing. SMN is oligomeric and forms a complex with Gemin2 (formerly SIP1), Gemin3 (a DEAD box RNA helicase), Gemin4, Gemin5 and Gemin6, as well as several spliceosomal snRNP proteins. The SMN complex plays an essential role in splicesomal snRNP assembly in the cytoplasm and is required for pre-mRNA splicing of the nucleus. The SMN complex is found in both the cytoplasm and the nucleus. The nuclear form is concentrated in subnuclear bodies called gems (gemini of the coiled bodies). Cytoplasmic SMN interacts with spliceosomal Sm proteins and facilitates their assembly onto U snRNAs, and nuclear SMN mediates recycling of pre-mRNA splicing factors. Nearly identical telomeric and centromeric forms of SMN encode the same protein; however, only mutations in the telomeric form are associated with the disease-state SMA. SMN is expresed in a wide variety of tissues including brain, kidney, liver, spinal cord and moderately in skeletal and cardiac muscle.

REFERENCES

1. Coovert, D., et al. 1997. The survival motor neuron protein in spinal muscular atrophy. Hum. Mol. Genet. 6: 1205-1214.
2. Fischer, U., et al. 1997. The SMN-SIP1 complex has an essential role in spliceosomal snRNP biogenesis. Cell 90: 1023-1029.

CHROMOSOMAL LOCATION

Genetic locus: SMN1 (human) mapping to 5q13.2.

PRODUCT

SMN 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 SMN shRNA Plasmid (h): sc-36510-SH and SMN shRNA (h) Lentiviral Particles: sc-36510-V as alternate gene silencing products.

For independent verification of SMN (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-36510A, sc-36510B and sc-36510C.

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

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

SMN (2B1): sc-32313 is recommended as a control antibody for monitoring of SMN 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 SMN gene expression knockdown using RT-PCR Primer: SMN (h)-PR: sc-36510-PR (20 µl, 455 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Gao, X., et al. 2012. Tudor staphylococcal nuclease (Tudor-SN) participates in small ribonucleoprotein (snRNP) assembly via interacting with symmetrically dimethylated Sm proteins. J. Biol. Chem. 287: 18130-18141.
2. Rodriguez-Muela, N., et al. 2018. Blocking p62-dependent SMN degradation ameliorates spinal muscular atrophy disease phenotypes. J. Clin. Invest. 128: 3008-3023.

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.