SANTA CRUZ BIOTECHNOLOGY, INC.

SRp75 siRNA (m): sc-44368

BACKGROUND

SRp75, also designated splicing factor, arginine/serine-rich 4 (SFRS4), is a splicing factor that can undergo phosphorylation. SRp75 is similar to other SR proteins, containing an N-terminal RNA recognition motif (RRM), a glycine-rich region, an internal region homologous to the RRM and a long 315 amino acid C-terminal serine/arginine-rich domain. SRp75, Pnn, SRm300 and SRrp130 components of spliceosome machinery can co-localize and co-immunoprecipitate with one another and exhibit speckled nuclear distribution that aligns with components of pre-mRNA splicing machinery. Alternative mRNA splicing plays an important role in development and differentiation; many transcripts are spliced differently in distinct cell types and tissues. Both constitutive and alternative splicing occurs on spliceosomes, which are complex particles composed of small nuclear ribonucleoproteins (snRNPs) and non-snRNP proteins. The SR family of non-snRNP splicing factors contain an RNA recognition motif and a serine- and arginine-rich (SR) domain. SR proteins are required at early stages of spliceosome assembly, have distinct but overlapping specificities for different pre-mRNAs and can alter splice site choice.

REFERENCES

1. Zahler, A.M., et al. 1993. Human SR proteins and isolation of a cDNA encoding SRp75. Mol. Cell. Biol. 13: 4023-4028.
2. ten Dam, G.B., et al. 1999. Alternative splicing of CD45 pre-mRNA is uniquely obedient to conditions in lymphoid cells. Biochim. Biophys. Acta 1446: 317-333.
3. Ko, B., et al. 2002. Identification of new poly(A) polymerase-inhibitory proteins capable of regulating pre-mRNA polyadenylation. J. Mol. Biol. 318: 1189-1206.
4. Zimowska, G., et al. 2003. Pinin/DRS/memA interacts with SRp75, SRm300 and SRrp130 in corneal epithelial cells. Invest. Ophthalmol. Vis. Sci. 44: 4715-4723.
5. Li, X., et al. 2005. New talents for an old acquaintance: the SR protein splicing factor ASF/SF2 functions in the maintenance of genome stability. Cell Cycle 4: 1706-1708.
6. Sanford, J.R., et al. 2005. Reversible phosphorylation differentially affects nuclear and cytoplasmic functions of splicing factor 2/alternative splicing factor. Proc. Natl. Acad. Sci. USA 102: 15042-15047.

CHROMOSOMAL LOCATION

Genetic locus: Srsf4 (mouse) mapping to 4 D2.3.

PRODUCT

SRp75 siRNA (m) 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 SRp75 shRNA Plasmid (m): sc-44368-SH and SRp75 shRNA (m) Lentiviral Particles: sc-44368-V as alternate gene silencing products.

For independent verification of SRp75 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44368A, sc-44368B and sc-44368C.

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

SRp75 siRNA (m) is recommended for the inhibition of SRp75 expression in mouse 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

SRp55 (16H3): sc-57954 is recommended as a control antibody for monitoring of SRp75 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 SRp75 gene expression knockdown using RT-PCR Primer: SRp75 (m)-PR: sc-44368-PR (20 µl). 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.