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splicing factor 1 shRNA (h) Lentiviral Particles: sc-44115-V

BACKGROUND

Mammalian splicing factor 1 (designated SF1, zinc finger protein 162, ZFM1, CW17R and mammalian branch point binding protein [mBBP]) specifically recognizes the seven-nucleotide branch point sequence located at 3' splice sites and participates in the assembly of early spliceosomal complexes. Splicing factor 1 functions as a transcriptional repressor and may control both proliferation and expression of pro-inflammatory gene products in smooth muscle cells. In addition, cytokine-induced downregulation of splicing factor 1 expression may contribute to the pathogenesis of hyperproliferative inflammatory diseases. The structure of splicing factor 1 contains a nuclear transport domain, a metal binding motif, and glutamine- and proline-rich regions. Human splicing factor 1 also exists as several different isoforms, H1-isoform and Bo-isoform, produced by alternative splicing events. The human splicing factor 1 gene is located on chromosome 11 close to the gene encoding Menin, the gene responsible for multiple endocrine neoplasia-type 1 (MEN1).

REFERENCES

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2. Kramer, A., et al. 1998. Diverse modes of alternative splicing of human splicing factor SF1 deduced from the exon-intron structure of the gene. *Gene* 211: 29-37.
3. Peled-Zehavi, H., et al. 2001. Recognition of RNA branch point sequences by the KH domain of splicing factor 1 (mammalian branch point binding protein) in a splicing factor complex. *Mol. Cell. Biol.* 21: 5232-5241.
4. Liu, Z., et al. 2001. Structural basis for recognition of the intron branch site RNA by splicing factor 1. *Science* 294: 1098-1102.
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CHROMOSOMAL LOCATION

Genetic locus: SF1 (human) mapping to 11q13.1.

PRODUCT

splicing factor 1 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 2 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see splicing factor 1 siRNA (h): sc-44115 and splicing factor 1 shRNA Plasmid (h): sc-44115-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

splicing factor 1 shRNA (h) Lentiviral Particles is recommended for the inhibition of splicing factor 1 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

splicing factor 1 (H-9): sc-398881 is recommended as a control antibody for monitoring of splicing factor 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgM-HRP: sc-2064 (dilution range: 1:500-1:5,000), TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgM-FITC: sc-2082 (dilution range: 1:100-1:400) or goat anti-mouse IgM-TR: sc-2983 (dilution range: 1:100-1:400) with UltraCruz[™] Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor splicing factor 1 gene expression knockdown using RT-PCR Primer: splicing factor 1 (h)-PR: sc-44115-PR (20 μ l, 589 bp). Annealing temperature for the primers should be $55-60^{\circ}$ C and the extension temperature should be $68-72^{\circ}$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

PROTOCOLS

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