SANTA CRUZ BIOTECHNOLOGY, INC.

SAP 114 siRNA (h): sc-62974

BACKGROUND

SAP 114 (spliceosome associated protein 114), also known as PRP21, PRPF21, SF3A120 or SF3A1, is a 793 amino acid ubiquitously expressed nuclear protein belonging to the SURP protein family and contains two SURP motif repeats and one ubiquitin-like domain. SAP 114 is a subunit of the SF3A splicing factor, a heterotrimeric complex comprised of three subunits that act in tandem to mediate the binding of U2 snRNP to the branchpoint sequence (BPS) in pre-mRNA. The SF3A complex is necessary for the conversion of 15S U2 snRNP into the active 17S protein that performs directly in pre-mRNA splicing events. Functioning as the first subunit of the complex, SAP 114 interacts with subunit 2 (SAP 62) and subunit 3 (SAP 61) via its SURP motifs. SAP 114 is also thought to be involved in the assembly of the E complex, a critical regulator of cell cycle progression from G_1 into S phase in mammalian cells.

REFERENCES

1. Chiara, M.D., et al. 1994. Specific protein-protein interactions between the essential mammalian spliceosome-associated proteins SAP 61 and SAP 114. Proc. Natl. Acad. Sci. USA 91: 6403-6407.
2. Seghezzi, W., et al. 1998. Cyclin E associates with components of the pre-mRNA splicing machinery in mammalian cells. Mol. Cell. Biol. 18: 4526-4536.
3. Das, R., et al. 2000. Functional association of U2 snRNP with the ATP-independent spliceosomal complex E. Mol. Cell 5: 779-787.
4. Will, C.L., et al. 2001. A novel U2 and U11/U12 snRNP protein that associates with the pre-mRNA branch site. EMBO J. 20: 4536-4546.
5. Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 605595. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
6. Jurica, M.S., et al. 2002. Purification and characterization of native spliceosomes suitable for three-dimensional structural analysis. RNA 8: 426-439.
7. Szabo, A., et al. 2004. Statistical modeling for selecting housekeeper genes. Genome Biol. 5: R59.
8. Rush, J., et al. 2005. Immunoaffinity profiling of tyrosine phosphorylation in cancer cells. Nat. Biotechnol. 23: 94-101.

CHROMOSOMAL LOCATION

Genetic locus: SF3A1 (human) mapping to 22q12.2.

PRODUCT

SAP 114 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 SAP 114 shRNA Plasmid (h): sc-62974-SH and SAP 114 shRNA (h) Lentiviral Particles: sc-62974-V as alternate gene silencing products.

For independent verification of SAP 114 (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-62974A, sc-62974B and sc-62974C.

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

SAP 114 siRNA (h) is recommended for the inhibition of SAP 114 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

SAP 114 (H-10): sc-515804 is recommended as a control antibody for monitoring of SAP 114 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 SAP 114 gene expression knockdown using RT-PCR Primer: SAP 114 (h)-PR: sc-62974-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.