

60 kDa Ro/SSA siRNA (h): sc-40918

BACKGROUND

Ro autoantigens are of clinical significance because antibodies directed against them are found in most patients with primary Sjogren syndrome, subacute cutaneous lupus erythematosus (SLE), neonatal lupus erythematosus, ANA-negative lupus erythematosus and systemic lupus erythematosus-like disease secondary to homozygous C2 or C4 complement deficiency. Ro/SSA is a ribonucleoprotein that binds to autoantibodies in 35 to 50% of patients with SLE and in up to 97% of patients with Sjogren syndrome. The Ro/SSA particle consists of a single immunoreactive protein noncovalently bound with one of four small RNA molecules. Most anti-Ro/SSA-positive sera have antibodies not only against the immunoreactive protein, but also against an Ro/SSA protein. The genes which encode the smaller and larger proteins map to human chromosomes 11p15.5 and 1q31, respectively. La/SSB is an autoimmune RNA-binding protein that plays a role in the transcription of RNA polymerase III was originally defined by its reactivity with autoantibodies from patients with Sjogren syndrome and SLE.

REFERENCES

- Chambers, J.C., et al. 1988. Genomic structure and amino acid sequence domains of the human La autoantigen. *J. Biol. Chem.* 263: 18043-18051.
- Itoh, K., et al. 1991. Protein heterogeneity in the human Ro/SSA ribonucleoproteins. The 52- and 60 kDa Ro/SSA autoantigens are encoded by separate genes. *J. Clin. Invest.* 87: 177-186.
- Frank, M.B., et al. 1993. The mapping of the human 52 kDa Ro/SSA autoantigen gene to human chromosome 11, and its polymorphisms. *Am. J. Hum. Genet.* 52: 183-191.

CHROMOSOMAL LOCATION

Genetic locus: TROVE2 (human) mapping to 1q31.2.

PRODUCT

60 kDa Ro/SSA 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see 60 kDa Ro/SSA shRNA Plasmid (h): sc-40918-SH and 60 kDa Ro/SSA shRNA (h) Lentiviral Particles: sc-40918-V as alternate gene silencing products.

For independent verification of 60 kDa Ro/SSA (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-40918A, sc-40918B and sc-40918C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

60 kDa Ro/SSA siRNA (h) is recommended for the inhibition of 60 kDa Ro/SSA 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 μ M in 66 μ l. 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

60 kDa Ro/SSA (AA-3): sc-100844 is recommended as a control antibody for monitoring of 60 kDa Ro/SSA 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 60 kDa Ro/SSA gene expression knockdown using RT-PCR Primer: 60 kDa Ro/SSA (h)-PR: sc-40918-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Pollak, A.J., et al. 2020. Gapmer antisense oligonucleotides targeting 5S ribosomal RNA can reduce mature 5S ribosomal RNA by two mechanisms. *Nucleic Acid Ther.* E-published.

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.