

60 kDa Ro/SSA siRNA (m): sc-40919

BACKGROUND

Ro autoantigens are of clinical significance because antibodies directed against them are found in most patients with primary Sjogren syndrome, sub-acute 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.
- Chan, E.K., et al. 1994. Human 60 kDa SSA/Ro ribonucleoprotein autoantigen gene (SSA2) localized to 1q31 by fluorescence *in situ* hybridization. *Genomics* 23: 298-300.
- Kurien, B.T., et al. 2001. Autoantibody to the leucine zipper region of 52 kDa Ro/SSA binds native 60 kDa Ro/SSA: identification of a tertiary epitope with components from 60 kDa Ro/SSA and 52 kDa Ro/SSA. *Scand. J. Immunol.* 53: 268-276.
- Hogg, J.R. and Collins, K. 2007. Human Y5 RNA specializes a Ro ribonucleoprotein for 5S ribosomal RNA quality control. *Genes Dev.* 21: 3067-3072.

CHROMOSOMAL LOCATION

Genetic locus: Trove2 (mouse) mapping to 1 F.

PRODUCT

60 kDa Ro/SSA 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see 60 kDa Ro/SSA shRNA Plasmid (m): sc-40919-SH and 60 kDa Ro/SSA shRNA (m) Lentiviral Particles: sc-40919-V as alternate gene silencing products.

For independent verification of 60 kDa Ro/SSA (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-40919A, sc-40919B and sc-40919C.

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 (m) is recommended for the inhibition of 60 kDa Ro/SSA 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 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 (m)-PR: sc-40919-PR (20 μ l). 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.