

EXOSC9 siRNA (h): sc-88907

BACKGROUND

The exosome is a multisubunit complex composed of several highly conserved subunits, some of which are 3' to 5' exoribonucleases. The complex is involved in a variety of cellular processes and is responsible for degrading unstable mRNAs that contain AU-rich (ARE) elements in their untranslated 3' region. EXOSC9 (exosome component 9), also called p5, p6, PM/ScI-75 or RRP45, is a component of the exosome complex and is required for processing of 7S pre-RNA to mature 5.8S rRNA. Located in the nucleus and cytoplasm, EXOSC9 is a ribonuclease that is involved in mRNA degradation, but does not contribute to stability of the exosome complex. Unlike most of the exosome subunits, EXOSC9 is thought to act both independently and as a member of the exosome, thus making it an essential part of ARE-mediated mRNA decay. EXOSC9 is proteolytically cleaved during apoptosis and is implicated in certain autoimmune diseases such as myositis and scleroderma. Four isoforms of this protein exist due to alternative splicing events.

REFERENCES

1. Brouwer, R., et al. 2002. Autoantibodies directed to novel components of the PM/ScI complex, the human exosome. *Arthritis Res.* 4: 134-138.
2. Mukherjee, D., et al. 2002. The mammalian exosome mediates the efficient degradation of mRNAs that contain AU-rich elements. *EMBO J.* 21: 165-174.
3. Raijmakers, R., et al. 2002. Protein-protein interactions between human exosome components support the assembly of RNase PH-type subunits into a six-membered PNPase-like ring. *J. Mol. Biol.* 323: 653-663.
4. Raijmakers, R., et al. 2003. The association of the human PM/ScI-75 autoantigen with the exosome is dependent on a newly identified N terminus. *J. Biol. Chem.* 278: 30698-30704.
5. Raijmakers, R., et al. 2004. PM-ScI-75 is the main autoantigen in patients with the polymyositis/scleroderma overlap syndrome. *Arthritis Rheum.* 50: 565-569.
6. Mahler, M., et al. 2005. Clinical evaluation of autoantibodies to a novel PM/ScI peptide antigen. *Arthritis Res. Ther.* 7: R704-R713.
7. Schilders, G., et al. 2007. Caspase-mediated cleavage of the exosome subunit PM/ScI-75 during apoptosis. *Arthritis Res. Ther.* 9: R12.

CHROMOSOMAL LOCATION

Genetic locus: EXOSC9 (human) mapping to 4q27.

PRODUCT

EXOSC9 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 EXOSC9 shRNA Plasmid (h): sc-88907-SH and EXOSC9 shRNA (h) Lentiviral Particles: sc-88907-V as alternate gene silencing products.

For independent verification of EXOSC9 (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-88907A, sc-88907B and sc-88907C.

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

EXOSC9 siRNA (h) is recommended for the inhibition of EXOSC9 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

EXOSC9 (D-6): sc-271815 is recommended as a control antibody for monitoring of EXOSC9 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 EXOSC9 gene expression knockdown using RT-PCR Primer: EXOSC9 (h)-PR: sc-88907-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.