

MsrA siRNA (m): sc-72127

BACKGROUND

Protein-bound methionine residues are among the most susceptible to oxidative stress by biological reactive oxygen species (ROS) such as H₂O₂, hydroxyl radicals and superoxide ions, which result in the formation of methionine sulfoxide, or Met(O). Methionine sulfoxide reductase (MsrA) is an enzyme that catalyzes the thioredoxin-dependent reduction of Met(O) residues in proteins and in methyl sulfoxide compounds. MsrA is an ubiquitously expressed protein which is found in organisms from yeast to man. Mammalian MsrA is most highly expressed in liver, kidney, macrophages, neutrophils, cerebellum and brain neurons. Oxidation of proteins by ROS is associated with oxidative stress and age-related diseases such as Alzheimer's disease. Recombinant mammalian MsrA retains enzymatic activity, and overexpression of the protein in yeast and human T cells increases their resistance to oxidative stress. Furthermore, MsrA activity decreases in all regions of the Alzheimer's disease brain. These findings indicate that MsrA plays an important role in protecting cells against oxidative damage and early cell death.

REFERENCES

1. Moskovitz, J., et al. 1996. Cloning the expression of a mammalian gene involved in the reduction of methionine sulfoxide residues in proteins. *Proc. Natl. Acad. Sci. USA* 93: 2095-2099.
2. Moskovitz, J., et al. 1996. Chromosomal localization of the mammalian peptide-methionine sulfoxide reductase gene and its differential expression in various tissues. *Proc. Natl. Acad. Sci. USA* 93: 3205-3208.
3. Moskovitz, J., et al. 1998. Overexpression of peptide-methionine sulfoxide reductase in *Sacharomyces cerevisiae* and human T cells provides them with high resistance to oxidative stress. *Proc. Natl. Acad. Sci. USA* 95: 14071-14075.
4. Kuschel, L., et al. 1999. Molecular cloning and functional expression of a human peptide methionine sulfoxide reductase (hMsrA). *FEBS Lett.* 456: 17-21.
5. Gabbita, S., et al. 1999. Decrease in peptide methionine sulfoxide reductase in Alzheimer's disease brain. *J. Neurochem.* 73: 1660-1666.
6. Lowther, W., et al. 2000. Structure and mechanism of peptide methionine sulfoxide reductase, an "anti-oxidation" enzyme. *Biochemistry* 39: 13307-13312.

CHROMOSOMAL LOCATION

Genetic locus: MsrA (mouse) mapping to 14 D1.

PRODUCT

MsrA 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 MsrA shRNA Plasmid (m): sc-72127-SH and MsrA shRNA (m) Lentiviral Particles: sc-72127-V as alternate gene silencing products.

For independent verification of MsrA (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-72127A, sc-72127B and sc-72127C.

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

MsrA siRNA (m) is recommended for the inhibition of MsrA 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

MsrA (1C8): sc-59619 is recommended as a control antibody for monitoring of MsrA 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor MsrA gene expression knockdown using RT-PCR Primer: MsrA (m)-PR: sc-72127-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.