SANTA CRUZ BIOTECHNOLOGY, INC.

MSR siRNA (m): sc-61079

BACKGROUND

The methylation of homocysteine to methionine (an essential amino acid in humans) is catalyzed by a cobalamin-dependent enzyme, methionine synthase (MS), for which MSR (methionine synthase reductase) acts as a chaperone protein. Over time, the cob(I)alamin cofactor of MS becomes oxidized to cob(II)alamin, thus inactivating MS. Regeneration of active MS requires reductive methylation catalyzed by MSR in which SAM is used as a methyl donor. MSR maintains MS activity at a 1:1 stoichiometric ratio. In the presence of MSR and NADPH, holoenzyme formation from apoMS and methylcobalamin is significantly heightened due to stabilization of apoMS in the presence of MSR. MSR is also able to decrease aquacobalamin to cob(II)alamin in the presence of NADPH, which stimulates conversion of apoMS and aquacobalamin to holoMS.

REFERENCES

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3. Olteanu, H., et al. 2002. Differences in the efficiency of reductive activation of methionine synthase and exogenous electron acceptors between the common polymorphic variants of human methionine synthase reductase. Biochemistry 41: 13378-13385.
4. Vaughn, J.D., et al. 2004. Methionine synthase reductase 66A→G polymorphism is associated with increased plasma homocysteine concentration when combined with the homozygous methylenetetrahydrofolate reductase 677C→T variant. J. Nutr. 134: 2985-2990.
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6. Zavadáková, P., et al. 2005. cblE type of homocystinuria due to methionine synthase reductase deficiency: functional correction by minigene expression. Hum. Mutat. 25: 239-247.
7. Zhang, Z., et al. 2005. Polymorphisms of methionine synthase and methionine synthase reductase and risk of squamous cell carcinoma of the head and neck: a case-control analysis. Cancer Epidemiol. Biomarkers Prev. 14: 1188-1193.
8. Ishikawa, H., et al. 2006. A polymorphism of the methionine synthase reductase gene increases chromosomal damage in peripheral lymphocytes in smokers. Mutat. Res. 599: 135-143.
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PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

CHROMOSOMAL LOCATION

Genetic locus: Mtrr (mouse) mapping to 13 B3.

PRODUCT

MSR 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MSR shRNA Plasmid (m): sc-61079-SH and MSR shRNA (m) Lentiviral Particles: sc-61079-V as alternate gene silencing products.

For independent verification of MSR (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-61079A, sc-61079B and sc-61079C.

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

MSR siRNA (m) is recommended for the inhibition of MSR 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor MSR gene expression knockdown using RT-PCR Primer: MSR (m)-PR: sc-61079-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.