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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

MTHFR siRNA (h): sc-43950

BACKGROUND

5,10-methylenetetrahydrofolate reductase (MTHFR) is an essential enzyme in the folate-dependent regulation of methionine and homocysteine concentrations. MTHFR catalyzes the reduction of 5,10-methylenetetrahydrofolate (methyleneTHF) to 5-methyltetrahydrofolate (methylTHF). MethylTHF, the predominate form of circulating folate, is the principle carbon donor for homocysteine methylation, a reaction that yields methionine. Folate prevents neural tube defects, and can lower homocysteine levels, suggesting that MTHFR function is important in preventing cardiovascular disease. Mutations at the MTHFR gene may influence vascular changes, predisposition to leukemia, coronary artery disease and hyperhomocysteinemia.

REFERENCES

- Homberger, A., et al. 2000. Genomic structure and transcript variants of the human methylenetetrahydrofolate reductase gene. *Eur. J. Hum. Genet.* 8: 725-729.
- Lucock, M. 2000. Folic acid: nutritional biochemistry, molecular biology, and role in disease processes. *Mol. Genet. Metab.* 71: 121-138.
- Gaughan, D.J., et al. 2000. The human and mouse methylenetetrahydrofolate reductase (MTHFR) genes: genomic organization, mRNA structure and linkage to the CLCN6 gene. *Gene* 257: 279-289.
- Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 236250. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
- LocusLink Report (LocusID: 4524). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: MTHFR (human) mapping to 1p36.22.

PRODUCT

MTHFR 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 MTHFR shRNA Plasmid (h): sc-43950-SH and MTHFR shRNA (h) Lentiviral Particles: sc-43950-V as alternate gene silencing products.

For independent verification of MTHFR (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-43950A, sc-43950B and sc-43950C.

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

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

MTHFR (5D3F3): sc-517229 is recommended as a control antibody for monitoring of MTHFR 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 MTHFR gene expression knockdown using RT-PCR Primer: MTHFR (h)-PR: sc-43950-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.