

THTR1 siRNA (m): sc-108022

BACKGROUND

Humans lack biosynthesis pathways for the micronutrients thiamine and folate, however, regulation of these vitamins is necessary for normal cellular function. The SLC19A gene family products mediate membrane transport of these molecules across the membrane to meet cellular requirements; in particular, two transporter proteins differentially import and export thiamine. In the liver as well as other tissues, THTR1 is responsible for the cellular accumulation, that is the import, of thiamine. Uptake depends on many factors, including sodium levels, pH, saturation of thiamine, presence of structural analogues such as oxythiamin and amprolium, as well as membrane transport inhibitors like amiloride. The gene encoding THTR1, SLC19A2, is regulated by GKLF, NF-1 and SP-1. Mutations of the SLC19A2 gene cause thiamine deficiency disorders such as thiamine-responsive megaloblastic anemia (TRMA) by interfering with either the functionality or intracellular targeting of THTR1.

REFERENCES

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3. Said, H.M., et al. 2003. Cellular and molecular aspects of thiamin uptake by human liver cells: studies with cultured Hep G2 cells. *Biochim. Biophys. Acta* 1567: 106-112.
4. Reidling, J.C., et al. 2003. *In vitro* and *in vivo* characterization of the minimal promoter region of human thiamin transporter SLC19A2. *Am. J. Physiol., Cell Physiol.* 285: C633-C641.
5. Baron, D., et al. 2003. Disruption of transport activity in a D93H mutant thiamine transporter 1, from a Rogers syndrome family. *Eur. J. Biochem.* 270: 4469-4477.
6. Oyewumi, M.O., et al. 2003. Specific association of thiamine-coated gadolinium nanoparticles with human breast cancer cells expressing thiamine transporters. *Bioconjug. Chem.* 14: 404-411.
7. Liu, X.Y., et al. 2003. Restoration of high-level transport activity by human reduced folate carrier/ThTr1 thiamine transporter chimaeras: role of the transmembrane domain 6/7 linker region in reduced folate carrier function. *Biochem. J.* 369: 31-37.

CHROMOSOMAL LOCATION

Genetic locus: Slc19a2 (mouse) mapping to 1 H2.2.

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.

PRODUCT

THTR1 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 THTR1 shRNA Plasmid (m): sc-108022-SH and THTR1 shRNA (m) Lentiviral Particles: sc-108022-V as alternate gene silencing products.

For independent verification of THTR1 (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-108022A, sc-108022B and sc-108022C.

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

THTR1 siRNA (m) is recommended for the inhibition of THTR1 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor THTR1 gene expression knockdown using RT-PCR Primer: THTR1 (m)-PR: sc-108022-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.