

TRMT6 siRNA (h): sc-76756

BACKGROUND

Transfer RNA (tRNA) modifications help regulate the efficiency of mRNA translation by maintaining the correct reading frames within mRNA. TRMT6 (tRNA methyltransferase 6), also known as GCD10 or TRM6, is a 497 amino acid protein that localizes to the nucleus and exists as a heterodimer with TRM61. Expressed in liver, brain, ovary and testis, TRMT6 functions as a substrate-binding subunit of tRNA and is thought to catalyze the formation of N(1)-methyladenine at position 58 in initiator methionyl-tRNA. TRMT6 exists as three alternatively spliced isoforms that, in response to DNA damage, may be phosphorylated by ATM or ATR. The gene encoding TRMT6 maps to human chromosome 20, which houses over 600 genes and comprises nearly 2% of the human genome.

REFERENCES

1. Hirosawa, M., et al. 1999. Characterization of cDNA clones selected by the GeneMark analysis from size-fractionated cDNA libraries from human brain. *DNA Res.* 6: 329-336.
2. Lai, C.H., et al. 2000. Identification of novel human genes evolutionarily conserved in *Caenorhabditis elegans* by comparative proteomics. *Genome Res.* 10: 703-713.
3. Ozanick, S., et al. 2005. The bipartite structure of the tRNA m1A58 methyltransferase from *S. cerevisiae* is conserved in humans. *RNA* 11: 1281-1290.
4. Ozanick, S.G., et al. 2007. Conserved amino acids in each subunit of the heterologous tRNA m1A58 Mtase from *Saccharomyces cerevisiae* contribute to tRNA binding. *Nucleic Acids Res.* 35: 6808-6819.
5. Matsuo, S., et al. 2007. ATM and ATR substrate analysis reveals extensive protein networks responsive to DNA damage. *Science* 316: 1160-1166.
6. Wang, X., et al. 2008. Degradation of hypomodified tRNA(iMet) *in vivo* involves RNA-dependent ATPase activity of the DEXH helicase Mtr4p. *RNA* 14: 107-116.

CHROMOSOMAL LOCATION

Genetic locus: TRMT6 (human) mapping to 20p12.3.

PRODUCT

TRMT6 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 TRMT6 shRNA Plasmid (h): sc-76756-SH and TRMT6 shRNA (h) Lentiviral Particles: sc-76756-V as alternate gene silencing products.

For independent verification of TRMT6 (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-76756A, sc-76756B and sc-76756C.

PROTOCOLS

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

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

TRMT6 siRNA (h) is recommended for the inhibition of TRMT6 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

TRMT6 (F-3): sc-271752 is recommended as a control antibody for monitoring of TRMT6 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 TRMT6 gene expression knockdown using RT-PCR Primer: TRMT6 (h)-PR: sc-76756-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.