

THP siRNA (m): sc-41065

BACKGROUND

Tamm-Horsfall glycoprotein (also referred to as uromodulin or THP) is the most abundant protein found in normal urine. THP is expressed on the luminal surface of the membrane with the glycosyl phosphatidylinositol (GPI) anchor and excreted in urine at a rate of 50-100 mg per day. THP, uropontin and nephrocalcin are the three known urinary glycoproteins that affect the formation of calcium-containing kidney stones. THP is synthesized by kidney epithelial cells and is believed to play important and diverse roles in the urinary system, including renal water balance, immunosuppression, urinary stone formation and inhibition of bacterial adhesion. THP is nontoxic and blocks early events required for normal T-cell proliferation *in vitro*. The gene which encodes THP and is a candidate gene for nephrolithiasis maps to human chromosome 16p12.3.

REFERENCES

1. Tamm, I. and Horsfall, F.L., Jr. 1952. A mucoprotein derived from human urine which reacts with influenza, mumps, and Newcastle disease viruses. *J. Exp. Med.* 95: 71-97.
2. Muchmore, A.V. and Decker, J.M. 1985. Uromodulin: a unique 85-kilodalton immunosuppressive glycoprotein isolated from urine of pregnant women. *Science* 229: 479-481.
3. Pook, M.A., et al. 1993. Localization of the Tamm-Horsfall glycoprotein (uromodulin) gene to chromosome 16p12.3-16p13.11. *Ann. Hum. Genet.* 57: 285-290.
4. Jeanpierre, C., et al. 1993. Chromosomal assignment of the uromodulin gene (UMOD) to 16p13.11. *Cytogenet. Cell Genet.* 62: 185-187.
5. Fukuoka, S. and Kobayashi, K. 2001. Analysis of the C-terminal structure of urinary Tamm-Horsfall protein reveals that the release of the glycosyl phosphatidylinositol-anchored counterpart from the kidney occurs by phenylalanine-specific proteolysis. *Biochem. Biophys. Res. Commun.* 289: 1044-1048.
6. Zhu, X., et al. 2002. Isolation of mouse THP gene promoter and demonstration of its kidney-specific activity in transgenic mice. *Am. J. Physiol. Renal Physiol.* 282: 608-617.
7. LocusLink Report (LocusID: 191845). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: Umod (mouse) mapping to 7 F2.

PRODUCT

THP 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 THP shRNA Plasmid (m): sc-41065-SH and THP shRNA (m) Lentiviral Particles: sc-41065-V as alternate gene silencing products.

For independent verification of THP (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-41065A, sc-41065B and sc-41065C.

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

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

THP (B-2): sc-271022 is recommended as a control antibody for monitoring of THP 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 MarkerTM 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 THP gene expression knockdown using RT-PCR Primer: THP (m)-PR: sc-41065-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.