

TPPP siRNA (h): sc-76720

BACKGROUND

Tubulin family members are globular proteins important in the assembly of microtubules. Microtubules are structural components that play important roles in mitosis, cytokinesis and vesicle transport. TPPP (tubulin polymerization-promoting protein), also known as p24 and p25, is a widely expressed 219 amino acid protein found in the perinuclear region of the cytoplasm. TPPP may form dimers and functions in polymerizing tubulin into double-walled tubules, polymorphic aggregates, or stabilized blocks. TPPP overexpression prevents formation of the mitotic spindle assembly and breakdown of the nuclear envelope. TPPP is phosphorylated by TPK II and is encoded by a gene that maps to human chromosome 5, which contains 181 million base pairs and comprises nearly 6% of the human genome.

REFERENCES

1. Libman, E., et al. 1981. Lipodystrophia diabetica. *Med. Pregl.* 34: 49-53.
2. Vincze, O., et al. 2006. Tubulin polymerization promoting proteins (TPPPs): members of a new family with distinct structures and functions. *Biochemistry* 45: 13818-13826.
3. Skjoerringe, T., et al. 2006. P25 α /Tubulin polymerization promoting protein expression by myelinating oligodendrocytes of the developing rat brain. *J. Neurochem.* 99: 333-342.
4. Preusser, M., et al. 2007. TPPP/p25 in brain tumours: expression in non-neoplastic oligodendrocytes but not in oligodendroglioma cells. *Acta Neuropathol.* 113: 213-215.
5. Gaddam, V. and Nagarajan, R. 2007. A new entry to polycyclic indole derivatives via intramolecular imino Diels-Alder reaction: observation of unexpected reaction. *J. Org. Chem.* 72: 3573-3576.
6. Dephoure, N., et al. 2008. A quantitative atlas of mitotic phosphorylation. *Proc. Natl. Acad. Sci. USA* 105: 10762-10767.

CHROMOSOMAL LOCATION

Genetic locus: TPPP (human) mapping to 5p15.33.

PRODUCT

TPPP siRNA (h) is a target-specific 19-25 nt siRNA 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 TPPP shRNA Plasmid (h): sc-76720-SH and TPPP shRNA (h) Lentiviral Particles: sc-76720-V as alternate gene silencing products.

For independent verification of TPPP (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-76720A, sc-76720B and sc-76720C.

PROTOCOLS

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

APPLICATIONS

TPPP siRNA (h) is recommended for the inhibition of TPPP expression in human cells.

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.

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

TPPP (A-6): sc-515819 is recommended as a control antibody for monitoring of TPPP 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 TPPP gene expression knockdown using RT-PCR Primer: TPPP (h)-PR: sc-76720-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.