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# TCP-1 $\epsilon$ siRNA (h): sc-43447

## BACKGROUND

The protein TCP-1 (t complex polypeptide 1) is a subunit of the heterooligomeric complex CCT (chaperonin containing TCP-1) present in the eukaryotic cytosol. The CCT of eukaryotic cytosol is composed of eight different subunit species, TCP-1  $\alpha$ ,  $\beta$ ,  $\gamma$ ,  $\delta$ ,  $\epsilon$ ,  $\zeta$ ,  $\eta$  and  $\theta$ , each encoded by a different gene. Two  $\zeta$  subunits have been described: TCP-1  $\zeta$  (also designated TCP-1  $\zeta$ 1) and TCP-1  $\zeta$ 2. TCP-1 subunits are proposed to have independent functions in folding its *in vivo* substrates, the actins and tubulins. TCP-1 was first identified in the mouse as relevant for tail-less and embryonic lethal phenotypes. Sequences homologous to TCP-1 have been isolated in several other species, and the yeast TCP-1 has been shown to encode a molecular chaperone for Actin and Tubulin. TCP-1 found in mammalian cells and yeast plays an important role in the folding of cytosolic proteins.

## REFERENCES

1. Ahnert, V., et al. 1996. Cucumber T complex protein. Molecular cloning, bacterial expression and characterization within a 22-S cytosolic complex in cotyledons and hypocotyls. *Eur. J. Biochem.* 235: 114-119.
2. Iijima, M., et al. 1998. A *Dictyostelium discoideum* homologue to TCP-1 is essential for growth and development. *Gene* 213: 101-106.
3. Ritco-Vonsovici, M., et al. 2000. Defining the eukaryotic cytosolic chaperonin-binding sites in human Tubulins. *J. Mol. Biol.* 304: 81-98.
4. Hynes, G.M. and Willison, K.R. 2000. Individual subunits of the eukaryotic cytosolic chaperonin mediate interactions with binding sites located on subdomains of  $\beta$ -Actin. *J. Biol. Chem.* 275: 18985-18994.
5. Campos, E.G., et al. 2000. Cloning of the chaperonin T complex polypeptide 1 gene from *Schistosoma mansoni* and studies of its expression levels under heat shock and oxidative stress. *Parasitol. Res.* 86: 253-258.
6. Yokota, S.I., et al. 2000. Upregulation of cytosolic chaperonin CCT subunits during recovery from chemical stress that causes accumulation of unfolded proteins. *Eur. J. Biochem.* 267: 1658-1664.

## CHROMOSOMAL LOCATION

Genetic locus: CCT5 (human) mapping to 5p15.2.

## PRODUCT

TCP-1  $\epsilon$  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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TCP-1  $\epsilon$  shRNA Plasmid (h): sc-43447-SH and TCP-1  $\epsilon$  shRNA (h) Lentiviral Particles: sc-43447-V as alternate gene silencing products.

For independent verification of TCP-1  $\epsilon$  (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-43447A, sc-43447B and sc-43447C.

## PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

TCP-1  $\epsilon$  siRNA (h) is recommended for the inhibition of TCP-1  $\epsilon$  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  $\mu$ M in 66  $\mu$ 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

TCP-1  $\epsilon$  (G-3): sc-376188 is recommended as a control antibody for monitoring of TCP-1  $\epsilon$  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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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 $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 TCP-1  $\epsilon$  gene expression knockdown using RT-PCR Primer: TCP-1  $\epsilon$  (h)-PR: sc-43447-PR (20  $\mu$ l, 554 bp). 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.