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SANTA CRUZ BIOTECHNOLOGY, INC.

TCP-1 η siRNA (h): sc-43449

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 α, β, γ, δ, ε, ζ, η and θ, each encoded by a different gene. Two ζ subunits have been described: TCP-1 ζ (also designated TCP-1 ζ1) and TCP-1 ζ2. 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 β-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.
7. Kafri, G., et al. 2001. Nested allosteric interactions in the cytoplasmic chaperonin containing TCP-1. Protein Sci. 10: 445-449.

CHROMOSOMAL LOCATION

Genetic locus: CCT7 (human) mapping to 2p13.2.

PRODUCT

TCP-1 η 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TCP-1 η shRNA Plasmid (h): sc-43449-SH and TCP-1 η shRNA (h) Lentiviral Particles: sc-43449-V as alternate gene silencing products.

For independent verification of TCP-1 η (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-43449A, sc-43449B and sc-43449C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

TCP-1 η siRNA (h) is recommended for the inhibition of TCP-1 η 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 µM in 66 µl. 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 η (A-8): sc-271951 is recommended as a control antibody for monitoring of TCP-1 η 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 η gene expression knockdown using RT-PCR Primer: TCP-1 η (h)-PR: sc-43449-PR (20 µl). 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.