

TBCK siRNA (h): sc-88942

BACKGROUND

TBCK (TBC domain-containing protein kinase-like protein) also known as TBCKL or MGC16169 in human and A630047E20Rik in mice, is an 893 amino acid protein belonging to the protein kinase superfamily. TBCK contains one protein kinase domain, a Rab-GAP TBC domain and one rhodanese domain. Four TBCK isoforms are produced by alternative splicing events, and the gene encoding TBCK maps to human chromosome 4q24. Chromosome 4 represents approximately 6% of the human genome and contains nearly 900 genes. Notably, the Huntingtin gene, which is found to encode an expanded glutamine tract in cases of Huntington's disease, is on chromosome 4. FGFR-3 is also encoded on chromosome 4 and has been associated with thanatophoric dwarfism, achondroplasia, Muenke syndrome and bladder cancer. Chromosome 4 is also tied to Ellis-van Creveld syndrome, methylmalonic acidemia and polycystic kidney disease.

REFERENCES

1. Dobson, C.M., et al. 2002. Identification of the gene responsible for the cblA complementation group of vitamin B12-responsive methylmalonic acidemia based on analysis of prokaryotic gene arrangements. *Proc. Natl. Acad. Sci. USA* 99: 15554-15559.
2. Cowan, C.M. and Raymond, L.A. 2006. Selective neuronal degeneration in Huntington's disease. *Curr. Top. Dev. Biol.* 75: 25-71.
3. Versteegh, F.G., et al. 2007. Growth hormone analysis and treatment in Ellis-van Creveld syndrome. *Am. J. Med. Genet. A* 143A: 2113-2121.
4. Doherty, E.S., et al. 2007. Muenke syndrome (FGFR3-related craniosynostosis): expansion of the phenotype and review of the literature. *Am. J. Med. Genet. A* 143A: 3204-3215.
5. Chandler, R.J., et al. 2007. Metabolic phenotype of methylmalonic acidemia in mice and humans: the role of skeletal muscle. *BMC Med. Genet.* 8: 64.
6. de Frutos, C.A., et al. 2007. Snail1 is a transcriptional effector of FGFR-3 signaling during chondrogenesis and achondroplasias. *Dev. Cell* 13: 872-883.

CHROMOSOMAL LOCATION

Genetic locus: TBCK (human) mapping to 4q24.

PRODUCT

TBCK 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 TBCK shRNA Plasmid (h): sc-88942-SH and TBCK shRNA (h) Lentiviral Particles: sc-88942-V as alternate gene silencing products.

For independent verification of TBCK (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-88942A, sc-88942B and sc-88942C.

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

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

TBCK (WW-8): sc-81865 is recommended as a control antibody for monitoring of TBCK 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 TBCK gene expression knockdown using RT-PCR Primer: TBCK (h)-PR: sc-88942-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.