

ε Tubulin siRNA (m): sc-43487

BACKGROUND

Tubulin is a major cytoskeleton component that has five distinct forms, designated α , β , γ , δ , and ϵ tubulin. α and β Tubulins form heterodimers, which multimerize to form a microtubule filament. There are five β Tubulin isoforms (β -I, β -II, β -III, β -IVa and β -IVb) that are expressed in mammalian tissues. β -I and β -IV are present throughout the cytosol, β -II is present in the nuclei and nucleoplasm, and β -III is a neuron-specific cytoskeletal protein. γ Tubulin forms the gammaosome, which is required for nucleating microtubule filaments at the centrosome. Both δ Tubulin and ϵ Tubulin are associated with the centrosome. δ Tubulin is a homologue of the *Chlamydomonas* δ Tubulin Uni3 and is found in association with the centrioles, whereas ϵ Tubulin localizes to the pericentriolar material. ϵ Tubulin exhibits a cell-cycle-specific pattern of localization, first associating with only the older of the centrosomes in a newly duplicated pair and later associating with both centrosomes.

REFERENCES

1. Weisenberg, R. 1981. Invited review: the role of nucleotide triphosphate in Actin and Tubulin assembly and function. *Cell Motil.* 1: 485-497.
2. Zheng, Y., Jung, M.K. and Oakley, B.R. 1991. γ Tubulin is present in *Drosophila melanogaster* and *Homo sapiens* and is associated with the centrosome. *Cell* 65: 817-823.
3. Burns, R.G. 1991. α , β , and γ Tubulins: sequence comparisons and structural constraints. *Cell Motil. Cytoskeleton* 20: 181-189.
4. Leask, A. and Stearns, T. 1998. Expression of amino- and carboxyl-terminal γ and α Tubulin mutants in cultured epithelial cells. *J. Biol. Chem.* 273: 2661-2668.
5. Luduena, R.F. 1998. Multiple forms of Tubulin: different gene products and covalent modifications. *Int. Rev. Cytol.* 178: 207-275.
6. Walss, C., Kreisberg, J.I. and Luduena, R.F. 1999. Presence of the β II iso-type of Tubulin in the nuclei of cultured mesangial cells from rat kidney. *Cell Motil. Cytoskeleton* 42: 274-284.
7. Modig, C., Olsson, P.E., Barasoain, I., de Ines, C., Andreu, J.M., Roach, M.C., Luduena, R.F. and Wallin, M. 1999. Identification of β 3 and β 4 Tubulin isoforms in cold-adapted microtubules from Atlantic cod (*Gadus morhua*): antibody mapping and cDNA sequencing. *Cell Motil. Cytoskeleton* 42: 315-330.
8. Woulfe, J. and Munoz, D. 2000. Tubulin immunoreactive neuronal intranuclear inclusions in the human brain. *Neuropathol. Appl. Neurobiol.* 26: 161-171.
9. Chang, P. and Stearns, T. 2000. δ Tubulin and ϵ Tubulin: two new human centrosomal tubulins reveal new aspects of centrosome structure and function. *Nat. Cell Biol.* 2: 30-35.

CHROMOSOMAL LOCATION

Genetic locus: Tuba1 (mouse) mapping to 10 B1.

PROTOCOLS

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

PRODUCT

ϵ Tubulin 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 ϵ Tubulin shRNA Plasmid (m): sc-43487-SH and ϵ Tubulin shRNA (m) Lentiviral Particles: sc-43487-V as alternate gene silencing products.

For independent verification of ϵ Tubulin (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-43487A, sc-43487B and sc-43487C.

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

ϵ Tubulin siRNA (m) is recommended for the inhibition of ϵ Tubulin 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ϵ Tubulin gene expression knockdown using RT-PCR Primer: ϵ Tubulin (m)-PR: sc-43487-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.