SANTA CRUZ BIOTECHNOLOGY, INC.

ε Tubulin siRNA (h): sc-43486

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 gammasome, 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. Burns, R.G. 1991. α-, β-, and γ-Tubulins: sequence comparisons and structural constraints. Cell Motil. Cytoskeleton 20: 181-189.
3. Zheng, Y., et al. 1991. γ Tubulin is present in *Drosophila melangaster* and *Homo sapiens* and is associated with the centrosome. Cell 65: 817-823.
4. Leask, A. and Stearns, T. 1998. Expression of amino- and carboxyl-terminal γ and β 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., et al. 1999. Presence of the β2 isotype of Tubulin in the nuclei of cultured mesangial cells from rat kidney. Cell Motil. Cytoskeleton 42: 274-284.
7. Modig, C., et al. 1999. Identification of β3 and β4 Tubulin isotypes 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: TUBE1 (human) mapping to 6q21.

PROTOCOLS

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

PRODUCT

ε Tubulin 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 ε Tubulin shRNA Plasmid (h): sc-43486-SH and ε Tubulin shRNA (h) Lentiviral Particles: sc-43486-V as alternate gene silencing products.

For independent verification of é Tubulin (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-43486A, sc-43486B and sc-43486C.

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

ε Tubulin siRNA (h) is recommended for the inhibition of ε Tubulin 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

ε Tubulin (5F3B7): sc-517236 is recommended as a control antibody for monitoring of ε Tubulin 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor é Tubulin gene expression knockdown using RT-PCR Primer: ε Tubulin (h)-PR: sc-43486-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.