SANTA CRUZ BIOTECHNOLOGY, INC.

GCP2 siRNA (m): sc-77387

BACKGROUND

The γ-Tubulin complex is composed of γ Tubulin and the γ-Tubulin complex-associated proteins GCP2, GCP3, GCP4, GCP5 and GCP6, all of which are essential components of microtubule organizing centers. γ-Tubulin complex components are localized to both the centrosome, where they are involved in microtubule nucleation, and to the cytoplasm, where they exist as soluble complexes that can be recruited to the centrosome as needed. Although the GCP proteins are related, they have distinct roles which contribute to the proper function of the γ-Tubulin complex. GCP2 (γ-Tubulin complex component 2), also known as TUBGCP2 or SPBC97 (spindle pole body protein Spc97 homolog), is a ubiquitously expressed 902 amino acid protein that localizes to the centrosome and is involved in microtubule nucleation.

REFERENCES

1. Murphy, S.M., et al. 1998. The mammalian γ-Tubulin complex contains homologues of the yeast spindle pole body components spc97p and spc98p. J. Cell Biol. 141: 663-674.
2. Fava, F., et al. 1999. Human 76p: a new member of the γ Tubulin-associated protein family. J. Cell Biol. 147: 857-868.
3. Hillman, R.T., et al. 2004. An unappreciated role for RNA surveillance. Genome Biol. 5: R8.
4. Rush, J., et al. 2005. Immunoaffinity profiling of tyrosine phosphorylation in cancer cells. Nat. Biotechnol. 23: 94-101.
5. Delgehyr, N., et al. 2005. Microtubule nucleation and anchoring at the centrosome are independent processes linked by Ninein function. J. Cell Sci. 118: 1565-1575.
6. Arbildua, J.J., et al. 2006. Fluorescence resonance energy transfer and molecular modeling studies on 4',6-diamidino-2-phenylindole (DAPI) complexes with Tubulin. Protein Sci. 15: 410-419.
7. Stirling, P.C., et al. 2006. PhLP3 modulates CCT-mediated Actin and Tubulin folding via ternary complexes with substrates. J. Biol. Chem. 281: 7012-7021.

CHROMOSOMAL LOCATION

Genetic locus: Tubgcp2 (mouse) mapping to 7 F4.

PRODUCT

GCP2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GCP2 shRNA Plasmid (m): sc-77387-SH and GCP2 shRNA (m) Lentiviral Particles: sc-77387-V as alternate gene silencing products.

For independent verification of GCP2 (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-77387A, sc-77387B and sc-77387C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

GCP2 siRNA (m) is recommended for the inhibition of GCP2 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 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

GCP2 (F-3): sc-377117 is recommended as a control antibody for monitoring of GCP2 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 GCP2 gene expression knockdown using RT-PCR Primer: GCP2 (m)-PR: sc-77387-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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