

C-Nap1 siRNA (h): sc-72669

BACKGROUND

C-Nap1 (centrosome-associated protein), also known as Centrosomal Nek2-associated protein 1 or CEP2 (centrosomal protein 2), is localized to the cytoplasm and exists in every human cell. C-Nap1 is expressed as three isoforms and is a 2,442 amino acid protein. C-Nap1 is a centrosomal protein found at the tips of parental centrioles and in basal bodies of cellular projections, such as cilia. C-Nap1 is associated with the interphase stage of the cell cycle, during which C-Nap1 functions in the cohesion of centrioles. The phosphorylation of C-Nap1 plays a role in regulating whether the centrosomes are associating or dissociating. Nek2 phosphorylates the C-terminal end of C-Nap1 during mitosis, an event which is thought to play a role in the dissociation of centrosomes. C-Nap1 reaccumulates in centrosomes at the end of cellular division. Some autoimmune diseases are characterized by the production of antibodies against C-Nap1, suggesting that C-Nap1 may be involved in the pathogenesis of autoimmune diseases.

REFERENCES

1. Fry, A.M., et al. 1998. C-Nap1, a novel centrosomal coiled-coil protein and candidate substrate of the cell cycle-regulated protein kinase Nek2. *J. Cell Biol.* 141: 1563-1574.
2. Mayor, T., et al. 2000. The centrosomal protein C-Nap1 is required for cell cycle-regulated centrosome cohesion. *J. Cell Biol.* 151: 837-846.
3. Mayor, T., et al. 2002. The mechanism regulating the dissociation of the centrosomal protein C-Nap1 from mitotic spindle poles. *J. Cell Sci.* 115: 3275-3284.
4. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 609689. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Yang, J., et al. 2006. Rootletin interacts with C-Nap1 and may function as a physical linker between the pair of centrioles/basal bodies in cells. *Mol. Biol. Cell* 17: 1033-1040.
6. Graser, S., et al. 2007. Cep68 and Cep215 (Cdk5rap2) are required for centrosome cohesion. *J. Cell Sci.* 120: 4321-4331.
7. Kim, K., et al. 2008. A novel function of CEP135 as a platform protein of C-NAP1 for its centriolar localization. *Exp. Cell Res.* 314: 3692-3700.

CHROMOSOMAL LOCATION

Genetic locus: CEP250 (human) mapping to 20q11.22.

PRODUCT

C-Nap1 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 C-Nap1 shRNA Plasmid (h): sc-72669-SH and C-Nap1 shRNA (h) Lentiviral Particles: sc-72669-V as alternate gene silencing products.

For independent verification of C-Nap1 (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-72669A, sc-72669B and sc-72669C.

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

C-Nap1 siRNA (h) is recommended for the inhibition of C-Nap1 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

C-Nap1 (F-7): sc-390540 is recommended as a control antibody for monitoring of C-Nap1 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 C-Nap1 gene expression knockdown using RT-PCR Primer: C-Nap1 (h)-PR: sc-72669-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.

PROTOCOLS

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