SANTA CRUZ BIOTECHNOLOGY, INC.

CEP290 siRNA (m): sc-72866

BACKGROUND

Centrosomes are the major microtubule-organizing centers of mammalian cells. They are composed of a centriole pair and surrounding microtubule-nucleating material, termed pericentriolar material (PCM). Bipolar mitotic spindle assembly relies on two intertwined processes: centriole duplication and centrosome maturation. Failure to properly orchestrate centrosome duplication and maturation is subsequently linked to spindle defects, which can result in aneuploidy and promote cancer progression. The CEP290 (centrosomal protein of 290 kDa) gene encodes a protein of 2,479 amino acids that activates CREB-2-mediated transcription. Specifically, CEP290 ensures the correct localization of ciliary and phototransduction proteins in retinal photoreceptor cells. Mutations in the CEP290 gene have been identified in several diseases, including Joubert syndrome type 5 (JBTS5), Senior-Løken syndrome type 6 (SLSN6), Leber congenital amaurosis type 10 (LCA10) and Meckel syndrome type 4 (MKS4).

REFERENCES

1. Lange, B.M., et al. 2000. Centriole duplication and maturation in animal cells. Curr. Top. Dev. Biol. 49: 235-249.
2. Palazzo, R.E., et al. 2000. Centrosome maturation. Curr. Top. Dev. Biol. 49: 449-470.
3. Pelletier, L., et al. 2006. Centriole assembly in *Caenorhabditis elegans.* Nature 444: 619-623.
4. Valente, E.M., et al. 2006. Mutations in CEP290, which encodes a centrosomal protein, cause pleiotropic forms of Joubert syndrome. Nat. Genet. 38: 623-625.
5. Sayer, J.A., et al. 2006. The centrosomal protein nephrocystin-6 is mutated in Joubert syndrome and activates transcription factor ATF-4. Nat. Genet. 38: 674-681.
6. Helou, J., et al. 2007. Mutation analysis of NPHP6/CEP290 in patients with Joubert syndrome and Senior-Løken syndrome. J. Med. Genet. 44: 657-663.

CHROMOSOMAL LOCATION

Genetic locus: Cep290 (mouse) mapping to 10 D1.

PRODUCT

CEP290 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 CEP290 shRNA Plasmid (m): sc-72866-SH and CEP290 shRNA (m) Lentiviral Particles: sc-72866-V as alternate gene silencing products.

For independent verification of CEP290 (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-72866A, sc-72866B and sc-72866C.

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 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

CEP290 siRNA (m) is recommended for the inhibition of CEP290 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

CEP290 (G-4): sc-390637 is recommended as a control antibody for monitoring of CEP290 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 CEP290 gene expression knockdown using RT-PCR Primer: CEP290 (m)-PR: sc-72866-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Jun, J.H., et al. 2022. Reduced expression of TAZ inhibits primary cilium formation in renal glomeruli. Exp. Mol. Med. 54: 169-179.

RESEARCH USE

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