SANTA CRUZ BIOTECHNOLOGY, INC.

CENPJ siRNA (m): sc-62091

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The Power to Question

BACKGROUND

CENPJ (CPAP) is a centromeric protein that plays a role in brain size. The protein contains five coiled-coil domains, the most C-terminal of which includes a leucine zipper motif. CENPJ also has several potential protein phosphorylation sites and a C-terminal domain containing 21 nonamer G-box repeats. The CENPJ gene is expressed in neuroepithelium during prenatal neurogenesis, and it localizes to the spindle poles of cells undergoing mitosis. This localization suggests that a centrosomal mechanism controls neuron number in the developing mammalian brain. Mutations in the CENPJ gene have been linked to the condition autosomal recessive primary microcephaly. A novel 4 bp deletion in the gene has been characterized as a cause of protein truncation leading to the condition.

REFERENCES

1. Hung, L.Y., et al. 2000. Protein 4.1 R-135 interacts with a novel centrosomal protein (CPAP) which is associated with the γ-tubulin complex. Mol. Cell. Biol. 20: 7813-7825.
2. Peng, B., et al. 2002. CPAP is a novel stat5-interacting cofactor that augments stat5-mediated transcriptional activity. Mol. Endocrinol. 16: 2019-2033.
3. Triebel, F., et al. 2003. LAG-3: a regulator of T-cell and DC responses and its use in therapeutic vaccination. Trends Immunol. 24: 619-622.
4. Hung, L.Y., et al. 2004. Identification of a novel microtubule-destabilizing motif in CPAP that binds to tubulin heterodimers and inhibits microtubule assembly. Mol. Biol. Cell 15: 2697-2706.
5. Bond, J., et al. 2005. A centrosomal mechanism involving CDK5RAP2 and CENPJ controls brain size. Nat. Genet. 37: 353-355.
6. Cho, J.H., et al. 2006. Depletion of CPAP by RNAi disrupts centrosome integrity and induces multipolar spindles. Biochem. Biophys. Res. Commun. 339: 742-747.

CHROMOSOMAL LOCATION

Genetic locus: Cenpj (mouse) mapping to 14 C3.

PRODUCT

CENPJ 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 CENPJ shRNA Plasmid (m): sc-62091-SH and CENPJ shRNA (m) Lentiviral Particles: sc-62091-V as alternate gene silencing products.

For independent verification of CENPJ (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-62091A, sc-62091B and sc-62091C.

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

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

CENPJ (E-12): sc-166113 is recommended as a control antibody for monitoring of CENPJ 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 CENPJ gene expression knockdown using RT-PCR Primer: CENPJ (m)-PR: sc-62091-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.