

CENP-C siRNA (h): sc-37559

BACKGROUND

A replicated chromosome includes two kinetochores that control chromosome segregation during mitosis. The evolutionarily conserved centromere protein-C, CENP-C, is a kinetochore assembly protein. CENP-C is located on the fibers of the kinetochore and constitutes a kinetochore organizing center that tightly associates with DNA. CENP-C is necessary for the formation of a functional centromere, which indicates that CENP-C is important for mitotic progression. In addition, CENP-C is lost from centromeres during herpes simplex virus 1 infection, causing substantial structural changes in the kinetochore, which suggests that the structure of CENP-C is regulated during the cell cycle.

REFERENCES

1. Rieder, C.L., et al. 1998. The vertebrate cell kinetochore and its roles during mitosis. *Trends Cell Biol.* 8: 310-318.
2. Fukagawa, T., et al. 1999. CENP-C is necessary but not sufficient to induce formation of a functional centromere. *EMBO J.* 18: 4196-4209.
3. Dawe, R.K., et al. 1999. A maize homolog of mammalian CENPC is a constitutive component of the inner kinetochore. *Plant Cell* 11: 1227-1238.
4. Sugimoto, K., et al. 1999. Visualization of prekinetochore locus on the centromeric region of highly extended chromatin fibers: does kinetochore auto-antigen CENP-C constitute a kinetochore organizing center? *Chromosome Res.* 7: 9-19.
5. Everett, R.D., et al. 1999. Specific destruction of kinetochore protein CENP-C and disruption of cell division by herpes simplex virus immediate-early protein Vmw110. *EMBO J.* 18: 1526-1538.
6. Choo, K.H. 2000. Centromerization. *Trends Cell Biol.* 10: 182-188.

CHROMOSOMAL LOCATION

Genetic locus: CENPC1 (human) mapping to 4q13.2.

PRODUCT

CENP-C 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 CENP-C shRNA Plasmid (h): sc-37559-SH and CENP-C shRNA (h) Lentiviral Particles: sc-37559-V as alternate gene silencing products.

For independent verification of CENP-C (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-37559A, sc-37559B and sc-37559C.

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

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

CENP-C (C-6): sc-166099 is recommended as a control antibody for monitoring of CENP-C 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 CENP-C gene expression knockdown using RT-PCR Primer: CENP-C (h)-PR: sc-37559-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.