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Plk siRNA (h2): sc-44322

BACKGROUND

Plk (for polo-like kinase) encodes a serine/threonine kinase that is closely related to polo and CDC5, genes that are required for passage through mitosis in *Drosophila* and *Saccharomyces*, respectively. Polo and CDC5 both code for proteins that are involved in regulating the function of the mitotic spindle. Plk protein accumulates in the cell during the S and G₂ phases of the cell cycle and both protein content and catalytic activity peak at the onset of mitosis, followed by a rapid reduction after mitosis. Plk expression is detectable in mitotically active tissues such as colon and placenta, as well as in tumors of various origins. It has also been suggested that Plk may serve as a marker of cell proliferation.

REFERENCES

1. Sunkel, C.E. and Glover, D.M. 1988. Polo, a mitotic mutant of *Drosophila* displaying abnormal spindle poles. *J. Cell Sci.* 89: 25-38.
2. Kitada, K., et al. 1993. A multicopy suppressor gene of the *Saccharomyces cerevisiae* G₁ cell cycle mutant gene DBF4 encodes a protein kinase and is identified as CDC5. *Mol. Cell. Biol.* 13: 4445-4457.
3. Lake, R.J. and Jelinek, W.R. 1993. Cell cycle- and terminal differentiation-associated regulation of the mouse mRNA encoding a conserved mitotic protein kinase. *Mol. Cell. Biol.* 13: 7793-7801.
4. Hamanaka, R., et al. 1994. Cloning and characterization of human and murine homologues of the *Drosophila* polo serine-threonine kinase. *Cell Growth Differ.* 5: 249-257.
5. Holtrich, U., et al. 1994. Induction and downregulation of Plk, a human serine/threonine kinase expressed in proliferating cells and tumors. *Proc. Natl. Acad. Sci. USA* 91: 1736-1740.
6. Golsteyn, R.M., et al. 1994. Cell cycle analysis and chromosomal localization of human Plk1, a putative homologue of the mitotic kinases *Drosophila* polo and *Saccharomyces cerevisiae* CDC5. *J. Cell Sci.* 107: 1509-1517.

CHROMOSOMAL LOCATION

Genetic locus: PLK1 (human) mapping to 16p12.2.

PRODUCT

Plk siRNA (h2) 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 Plk shRNA Plasmid (h2): sc-44322-SH and Plk shRNA (h2) Lentiviral Particles: sc-44322-V as alternate gene silencing products.

For independent verification of Plk (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44322A, sc-44322B and sc-44322C.

PROTOCOLS

See our web site at www.scbt.com or our catalog 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 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

Plk siRNA (h2) is recommended for the inhibition of Plk 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

Plk (F-8): sc-17783 is recommended as a control antibody for monitoring of Plk 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Plk gene expression knockdown using RT-PCR Primer: Plk (h2)-PR: sc-44322-PR (20 μ l, 432 bp). 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.