Cdk9 siRNA (m): sc-35050



The Power to Question

BACKGROUND

A family of proteins designated cyclin dependent kinases (Cdks) are critical regulators of cell cycle progression. Cdk family members, including Cdc2 p34, Cdk1–9, PISSLRE, KKIALRE, PITSLRE and PCTAIRE 1–3 are constitutively expressed throughout the cell cycle. Cdc2 p34 activity peaks during mitosis and Cdk2 activity rises in late G_1 or early S phase. Cdk4 and Cdk6 are critically involved in G_1 to S phase progression. The functions of Cdk3, Cdk5, PISSLRE, KKIALRE and PCTAIRE 1–3 are less well defined. Cdk9 (also designated PITALRE) has been shown to specifically phosphorylate the retinoblastoma protein. The more recently cloned Drosophila protein, P-TEFb, is thought to be the homolog of mammalian PITALRE. P-TEFb has been shown to be required for HIV Tat transcriptional activation.

REFERENCES

- Rosenblatt, J., et al. 1992. Human cyclin-dependent kinase 2 is activated during the S and G₂ phases of the cell cycle and associates with cyclin A. Proc. Natl. Acad. Sci. USA 89: 2824-2828.
- 2. Okuda, T., et al. 1992. PCTAIRE-1 and PCTAIRE-3, two members of a novel Cdc2/Cdc28-related protein kinase gene family. Oncogene 7: 2249-2258.
- Grana, X., et al. 1994. PITALRE, a nuclear Cdc2-related protein kinase that phosphorylates the retinoblastoma protein *in vitro*. Proc. Natl. Acad. Sci. USA 91: 3834-3838.
- MacLachlan, T.K., et al. 1995. Cyclins, cyclin-dependent kinases and Cdk inhibitors: implications in cell cycle control and cancer. Crit. Rev. Eukaryot. Gene Expr. 5: 127-156.

CHROMOSOMAL LOCATION

Genetic locus: Cdk9 (mouse) mapping to 2 B.

PRODUCT

Cdk9 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 Cdk9 shRNA Plasmid (m): sc-35050-SH and Cdk9 shRNA (m) Lentiviral Particles: sc-35050-V as alternate gene silencing products.

For independent verification of Cdk9 (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-35050A, sc-35050B and sc-35050C.

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

Cdk9 siRNA (m) is recommended for the inhibition of Cdk9 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

Cdk9 (D-7): sc-13130 is recommended as a control antibody for monitoring of Cdk9 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz* Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 Cdk9 gene expression knockdown using RT-PCR Primer: Cdk9 (m)-PR: sc-35050-PR (20 μ l, 578 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

 Sharma, M., et al. 2007. Regulation of transcript elongation through cooperative and ordered recruitment of cofactors. J. Biol. Chem. 282: 20887-20896.

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.

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