SANTA CRUZ BIOTECHNOLOGY, INC.

cyclin K siRNA (m): sc-142657

BACKGROUND

Positive transcription elongation factor b (P-TEFb) complexes are crucial for allowing the elongation of RNA by RNA polymerase II (RNAPII). These complexes are able to phosphorylate the carboxyl-terminal domain of the largest RNAPII subunit. P-TEFb complexes are made up of a catalytic subunit, cyclin dependent kinase 9 (Cdk9), and one of the regulatory cyclins, CycT1, CycT2a, CycT2b or cyclin K. Specifically, cyclin K forms an active P-TEFb complex with Cdk9. This complex promotes transcription by phosphorylating the carboxyl-terminal domain of RNAPII which allows the elongation of transcription to proceed. Cyclin K is ubiquitously expressed in adult mouse and human tissues, with highest levels expressed in the developing germ cells of adult testis and ovaries. Cyclin K is also present in Hep G2 cells. The cyclin K gene encodes a 357 amino acid protein and maps to human chromosome 14q32.

REFERENCES

1. Edwards, M.C., et al. 1998. Human cyclin K, a novel RNA polymerase II-associated cyclin possessing both carboxy-terminal domain kinase and Cdk-activating kinase activity. Mol. Cell. Biol. 7: 4291-4300.
2. Fu, T.J., et al. 1999. Cyclin K functions as a Cdk9 regulatory subunit and participates in RNA polymerase II transcription. J. Biol. Chem. 274: 34527-34530.
3. Lin, X., et al. 2002. P-TEFb containing cyclin K and Cdk9 can activate transcription via RNA. J. Biol. Chem. 277: 16873-16878.
4. Mori, T., et al. 2002. Cyclin K as a direct transcriptional target of the p53 tumor suppressor. Neoplasia 4: 268-274.
5. Lundquist, A., et al. 2003. Kaposi sarcoma-associated viral cyclin K overrides cell growth inhibition mediated by Oncostatin M through Stat3 inhibition. Blood 101: 4070-4077.
6. SWISS-PROT/TrEMBL (O75909). World Wide Web URL: http://www.expasy.ch/sprot/sprot-top.html.

CHROMOSOMAL LOCATION

Genetic locus: Ccnk (mouse) mapping to 12 F1.

PRODUCT

cyclin K 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 cyclin K shRNA Plasmid (m): sc-142657-SH and cyclin K shRNA (m) Lentiviral Particles: sc-142657-V as alternate gene silencing products.

For independent verification of cyclin K (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-142657A, sc-142657B and sc-142657C.

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

cyclin K siRNA (m) is recommended for the inhibition of cyclin K 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

cyclin K (G-11): sc-376371 is recommended as a control antibody for monitoring of cyclin K 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 cyclin K gene expression knockdown using RT-PCR Primer: cyclin K (m)-PR: sc-142657-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.