

Cdk8 siRNA (h): sc-29267

BACKGROUND

Cell cycle progression is controlled in part by a family of cyclin proteins and cyclin dependent kinases (Cdks). Cdk proteins work in concert with the cyclins to phosphorylate key substrates involved in each phase of cell cycle progression. Another family of proteins, Cdk inhibitors, also plays a role in regulating cell cycle by binding to cyclin-Cdk complexes and modulating their activity. Several Cdk proteins have been identified, including Cdk2-Cdk8, PCTAIRE-1-3, PITSLRE and PITSRLRE. Large complexes containing Cdk8, cyclin C and the large subunit of RNA polymerase II have been identified. Cdk8 is thought to regulate RNA polymerase II function in conjunction with cyclin C. Cdk8 has been demonstrated to function as a transcriptional activator when fused to the DNA binding domain of GAL4.

CHROMOSOMAL LOCATION

Genetic locus: CDK8 (human) mapping to 13q12.13.

PRODUCT

Cdk8 siRNA (h) is a pool of 4 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 Cdk8 shRNA Plasmid (h): sc-29267-SH and Cdk8 shRNA (h) Lentiviral Particles: sc-29267-V as alternate gene silencing products.

For independent verification of Cdk8 (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-29267A, sc-29267B, sc-29267C and sc-29267D.

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

Cdk8 siRNA (h) is recommended for the inhibition of Cdk8 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

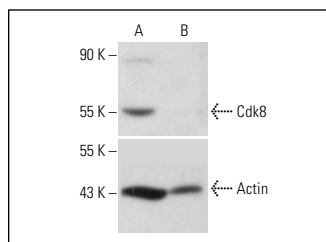
Cdk8 (D-9): sc-13155 is recommended as a control antibody for monitoring of Cdk8 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

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.

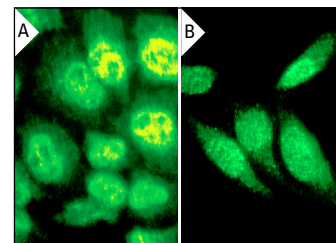
RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Cdk8 gene expression knockdown using RT-PCR Primer: Cdk8 (h)-PR: sc-29267-PR (20 μ l, 587 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

DATA



Cdk8 siRNA (h): sc-29267. Western blot analysis of Cdk8 expression in non-transfected control (A) and Cdk8 siRNA transfected (B) HeLa cells. Blot probed with Cdk8 (D-9): sc-13155. Actin (I-19): sc-1616 used as specificity and loading control.



Cdk8 siRNA (h): sc-29267. Immunofluorescence staining of methanol-fixed, control HeLa (A) and Cdk8 siRNA silenced HeLa (B) cells showing diminished nuclear staining in the siRNA silenced cells. Cells probed with Cdk8 (C-19): sc-1521

SELECT PRODUCT CITATIONS

- Brägelmann, J., et al. 2017. Pan-cancer analysis of the mediator complex transcriptome identifies Cdk19 and Cdk8 as therapeutic targets in advanced prostate cancer. *Clin. Cancer Res.* 23: 1829-1840.
- Zhang, J.F., et al. 2018. MicroRNA-770 affects proliferation and cell cycle transition by directly targeting Cdk8 in glioma. *Cancer Cell Int.* 18: 195.

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.