

CLK4 siRNA (h): sc-105214

BACKGROUND

The phosphorylation and dephosphorylation of proteins on serine and threonine residues is an essential means of regulating a broad range of cellular functions in eukaryotes, including cell division, homeostasis and apoptosis. A group of proteins that are intimately involved in this process are the serine/threonine (Ser/Thr) protein kinases. CLK4 (CDC-like kinase 4) is a 481 amino acid nuclear protein that contains one protein kinase domain and is a member of the Ser/Thr protein kinase family. Expressed in brain, liver, kidney, heart and muscle, CLK4 catalyzes the ATP-dependent phosphorylation of serine- and arginine-rich (SR) proteins within the spliceosomal complex and is thought to regulate the ability of SR proteins to control RNA splicing. CLK4 shares 97% sequence homology with its mouse counterpart, suggesting a highly conserved function between species.

REFERENCES

1. Hanks, S.K., et al. 1988. The protein kinase family: conserved features and deduced phylogeny of the catalytic domains. *Science* 241: 42-52.
2. Hanks, S.K., et al. 1991. Protein kinase catalytic domain sequence database: identification of conserved features of primary structure and classification of family members. *Methods Enzymol.* 200: 38-62.
3. Schultz, J., et al. 2001. Molecular characterization of a cDNA encoding functional human CLK4 kinase and localization to chromosome 5q35 [correction of 4q35]. *Genomics* 71: 368-370.
4. Katsu, R., et al. 2002. Novel SR-rich-related protein clasp specifically interacts with inactivated Clk4 and induces the exon EB inclusion of Clk. *J. Biol. Chem.* 277: 44220-44228.

CHROMOSOMAL LOCATION

Genetic locus: CLK4 (human) mapping to 5q35.3.

PRODUCT

CLK4 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 CLK4 shRNA Plasmid (h): sc-105214-SH and CLK4 shRNA (h) Lentiviral Particles: sc-105214-V as alternate gene silencing products.

For independent verification of CLK4 (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-105214A, sc-105214B and sc-105214C.

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

CLK4 siRNA (h) is recommended for the inhibition of CLK4 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

CLK1/4 (A-4): sc-515307 is recommended as a control antibody for monitoring of CLK4 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 CLK4 gene expression knockdown using RT-PCR Primer: CLK4 (h)-PR: sc-105214-PR (20 μ l, 547 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Petsalaki, E. and Zachos, G. 2016. Clks 1, 2 and 4 prevent chromatin breakage by regulating the Aurora B-dependent abscission checkpoint. *Nat. Commun.* 7: 11451.

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.