

CLK3 siRNA (h): sc-72925

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. CLK3 (CDC-like kinase 3), also known as PHCLK3, is a 638 amino acid nuclear and cytoplasmic protein that belongs to the Ser/Thr protein kinase family. Functioning as a dual-specificity kinase, CLK3 catalyzes the ATP-dependent phosphorylation of arginine- and serine-rich (SR) splicing factor proteins, thereby regulating both their function and their intranuclear distribution. Via its enzymatic activity, CLK3 is thought to be one of several members of a network of regulatory proteins that control RNA splicing events. Four isoforms of CLK3 exist due to alternative splicing.

REFERENCES

1. Becker, W., et al. 1996. cDNA cloning and characterization of rat Clk3, a LAMMER kinase predominately expressed in testis. *Biochim. Biophys. Acta* 1312: 63-67.
2. Duncan, P.L., et al. 1998. The Clk2 and Clk3 dual-specificity protein kinases regulate the intranuclear distribution of SR proteins and influence pre-mRNA splicing. *Exp. Cell Res.* 241: 300-308.
3. Menegay, H., et al. 1999. The dual specificity protein kinase CLK3 is abundantly expressed in mature mouse spermatozoa. *Exp. Cell Res.* 253: 463-473.
4. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 602990. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. García-Sacristán, A., et al. 2005. Protein kinase clk/STY is differentially regulated during erythroleukemia cell differentiation: a bias toward the skipped splice variant characterizes postcommitment stages. *Cell Res.* 15: 495-503.

CHROMOSOMAL LOCATION

Genetic locus: CLK3 (human) mapping to 15q24.1.

PRODUCT

CLK3 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 CLK3 shRNA Plasmid (h): sc-72925-SH and CLK3 shRNA (h) Lentiviral Particles: sc-72925-V as alternate gene silencing products.

For independent verification of CLK3 (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-72925A, sc-72925B and sc-72925C.

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 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

CLK3 siRNA (h) is recommended for the inhibition of CLK3 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

CLK3 (D-10): sc-365225 is recommended as a control antibody for monitoring of CLK3 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 CLK3 gene expression knockdown using RT-PCR Primer: CLK3 (h)-PR: sc-72925-PR (20 μ l). 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.