



dCK siRNA (m): sc-60510

BACKGROUND

The dCK/dGK family of proteins includes four different deoxyribonucleoside kinases including the cytoplasmic (TK1) and mitochondrial (TK2) thymidine kinases, and the deoxycytidine (dCK) and deoxyguanosine (dGK) kinases. Deoxyribonucleoside kinases catalyze the 5'-phosphorylation of 2'-deoxyribonucleosides with nucleoside triphosphates (NTPs) as phosphate donors. The dCK enzyme is associated with drug resistance and sensitivity, as both dCK and TK2 phosphorylate several antiviral and chemotherapeutic nucleoside analogs. Deficiency of dCK activity corresponds with resistance to antiviral and chemotherapeutic agents. dCK and TK1 localize to the cytosol, whereas dGK and TK2 localize to the mitochondria. These deoxyribonucleoside kinases are most abundantly expressed in muscle, brain and liver.

REFERENCES

1. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 125450. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
2. Galmarini, C.M., et al. 2005. The prognostic value of cN-II and cN-III enzymes in adult acute myeloid leukemia. *Haematologica* 90: 1699-701.
3. Bergman, A.M., et al. 2005. *In vivo* induction of resistance to gemcitabine results in increased expression of ribonucleotide reductase subunit M1 as the major determinant. *Cancer Res.* 65: 9510-9516.
4. Karbownik, M., et al. 2005. Increased expression of mRNA specific for thymidine kinase, deoxycytidine kinase or thymidine phosphorylase in human papillary thyroid carcinoma. *Cancer Lett.* 225: 267-273.
5. Hubeek, I., et al. 2005. Immunocytochemical detection of deoxycytidine kinase in haematological malignancies and solid tumours. *J. Clin. Pathol.* 58: 695-699.
6. Smal, C., et al. 2006. Identification of *in vivo* phosphorylation sites on human deoxycytidine kinase. Role of Ser-74 in the control of enzyme activity. *J. Biol. Chem.* 281: 4887-4893.

CHROMOSOMAL LOCATION

Genetic locus: Dck (mouse) mapping to 5 E1.

PRODUCT

dCK 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see dCK shRNA Plasmid (m): sc-60510-SH and dCK shRNA (m) Lentiviral Particles: sc-60510-V as alternate gene silencing products.

For independent verification of dCK (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-60510A, sc-60510B and sc-60510C.

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

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

dCK (H-3): sc-393099 is recommended as a control antibody for monitoring of dCK 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 dCK gene expression knockdown using RT-PCR Primer: dCK (m)-PR: sc-60510-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.