

DMPK siRNA (h): sc-38993

BACKGROUND

Myotonic dystrophy protein kinase (DMPK) is a multi-domain protein kinase found in muscle that is activated in response to G protein second messengers and proteolysis. DMPK is implicated in myotonic muscular dystrophy (DM), an autosomal dominant-inherited disorder that predominately affects skeletal and cardiac muscle and causes defects in cardiac conduction. DM arises through expansion of CTG repeats in the 3'-UTR of the DMPK gene. Mutant DMPK transcripts with an extended region of CUG repeats are retained in the nucleus. These transcripts also influence the expression of the DM locus-associated homeodomain protein (DMAHP)/SIX5, to mediate in part the DM phenotype. Other substrates for DMPK include myogenin, L-type calcium channels, and Phospholemman (PLM).

REFERENCES

1. Roberts, R., et al. 1997. Altered phosphorylation and intracellular distribution of a (CUG)_n triplet repeat RNA-binding protein in patients with myotonic dystrophy and in myotonin protein kinase knockout mice. *Proc. Natl. Acad. Sci. USA* 94: 13221-13226.
2. Berul, C.I., et al. 1999. DMPK dosage alterations result in atrioventricular conduction abnormalities in a mouse myotonic dystrophy model. *J. Clin. Invest.* 103: R1-R7.
3. Mounsey, J.P., et al. 2000. Phospholemman is a substrate for myotonic dystrophy protein kinase. *J. Biol. Chem.* 275: 23362-23367.
4. Bush, E.W., et al. 2000. Myotonic dystrophy protein kinase domains mediate localization, oligomerization, novel catalytic activity, and autoinhibition. *Biochemistry* 39: 8480-8490.
5. Mankodi, A., et al. 2000. Myotonic dystrophy in transgenic mice expressing an expanded CUG repeat. *Science* 289: 1769-1773.
6. Inukai, A., et al. 2000. Reduced expression of DMAHP/SIX5 gene in myotonic dystrophy muscle. *Muscle Nerve* 23: 1421-1426.

CHROMOSOMAL LOCATION

Genetic locus: DMPK (human) mapping to 19q13.32.

PRODUCT

DMPK 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 DMPK shRNA Plasmid (h): sc-38993-SH and DMPK shRNA (h) Lentiviral Particles: sc-38993-V as alternate gene silencing products.

For independent verification of DMPK (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-38993A, sc-38993B and sc-38993C.

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

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

DMPK (9-RY26): sc-134319 is recommended as a control antibody for monitoring of DMPK 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 DMPK gene expression knockdown using RT-PCR Primer: DMPK (h)-PR: sc-38993-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.