

MLKL siRNA (h): sc-93430

BACKGROUND

MLKL (mixed lineage kinase domain-like) is a 471 amino acid protein that contains one protein kinase domain which is thought to be catalytically inactive. The gene encoding MLKL maps to chromosome 16q23.1 and is expressed as two isoforms which are produced by alternative splicing events. Chromosome 16, which is associated with a variety of genetic disorders, encodes over 900 genes and comprises nearly 3% of the human genome. The GAN gene is located on chromosome 16q23.2 and, with mutation, may lead to giant axonal neuropathy, a nervous system disorder characterized by increasing malfunction with growth. The rare disorder Rubinstein-Taybi syndrome is associated with chromosome 16, as is Crohn's disease, which is a gastrointestinal inflammatory condition.

REFERENCES

- Gilbert, F. 1999. Disease genes and chromosomes: disease maps of the human genome. *Chromosome 16. Genet. Test.* 3: 243-254.
- Manning, G., et al. 2002. The protein kinase complement of the human genome. *Science* 298: 1912-1934.
- Coupry, I., et al. 2004. Analysis of CBP (CREBBP) gene deletions in Rubinstein-Taybi syndrome patients using real-time quantitative PCR. *Hum. Mutat.* 23: 278-284.

CHROMOSOMAL LOCATION

Genetic locus: MLKL (human) mapping to 16q23.1.

PRODUCT

MLKL 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 MLKL shRNA Plasmid (h): sc-93430-SH and MLKL shRNA (h) Lentiviral Particles: sc-93430-V as alternate gene silencing products.

For independent verification of MLKL (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-93430A, sc-93430B and sc-93430C.

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

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

MLKL (3B2): sc-293201 is recommended as a control antibody for monitoring of MLKL 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 MLKL gene expression knockdown using RT-PCR Primer: MLKL (h)-PR: sc-93430-PR (20 μ l, 576 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Miki, Y., et al. 2015. Photodynamic therapy using talaporfin sodium induces concentration-dependent programmed necroptosis in human glioblastoma T98G cells. *Lasers Med. Sci.* 30: 1739-1745.
- Kosic, M., et al. 2016. Synergistic anticancer action of lysosomal membrane permeabilization and glycolysis inhibition. *J. Biol. Chem.* 291: 22936-22948.
- Kitur, K., et al. 2016. Necroptosis promotes *Staphylococcus aureus* clearance by inhibiting excessive inflammatory signaling. *Cell Rep.* 16: 2219-2230.
- Ma, C., et al. 2022. Calycosin ameliorates atherosclerosis by enhancing autophagy via regulating the interaction between KLF2 and MLKL in apoE^{-/-} mice. *Br. J. Pharmacol.* 179: 252-269.

RESEARCH USE

For research use only, not for use in diagnostic procedures.