

Misato siRNA (h): sc-62619

BACKGROUND

Drosophila melanogaster is a proven and effective model for studying developmental and cellular processes common to higher eukaryotes. Approximately 13,600 genes have been elucidated from more than 120 megabases of euchromatin, and they are organized among the chromosomes 2, 3, 4, X and Y, with the Y chromosome being predominately heterochromatic. *Drosophila* genes can be categorized based on the type of protein for which they encode and are represented by six major classifications, which include intracellular signaling proteins, transmembrane proteins, RNA binding proteins, secreted factors, transcription regulators (basic helix-loop-helix, homeodomain containing, zinc finger containing, and chromatin associated) or other functional proteins. The Misato gene encodes a protein that contains a mixture of peptide motifs found in α , β and γ Tubulins, as well as a motif related to part of the Myosin heavy chain proteins. Null mutations at the Misato locus of *Drosophila melanogaster* are associated with irregular chromosomal segregation at cell division and result in larvae that have reduced levels of imaginal disk tissue, a reduction in brain size and die during the larval stage of development.

REFERENCES

1. Miklos, G.L., Yamamoto, M., Burns, R.G. and Maleszka, R. 1997. An essential cell division gene of *Drosophila*, absent from *Saccharomyces*, encodes an unusual protein with Tubulin-like and Myosin-like peptide motifs. *Proc. Natl. Acad. Sci. USA* 94: 5189-5194.
2. Nogales, E., Wolf, S.G. and Downing, K.H. 1998. Structure of the α/β Tubulin dimer by electron crystallography. *Nature* 391: 199-203.
3. Adams, M.D., Celniker, S.E., Holt, R.A., Evans, C.A., Gocayne, J.D., Amanatides, P., Scherer, S.E., Li, P.W., Hoskins, R.A., Galle, R.F., George, R.A., Lewis, S.E., Richards, S., Ashburner, M., Henderson, S.N., Sutton, G.G., Wortman, J.R., et al. 2000. The genome sequence of *Drosophila melanogaster*. *Science* 287: 2185-2195.
4. LocusLink Report (LocusID: 33119). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: MSTO1 (human) mapping to 1q22.

PRODUCT

Misato 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 Misato shRNA Plasmid (h): sc-62619-SH and Misato shRNA (h) Lentiviral Particles: sc-62619-V as alternate gene silencing products.

For independent verification of Misato (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-62619A, sc-62619B and sc-62619C.

RESEARCH USE

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

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

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

Misato (E-10): sc-390638 is recommended as a control antibody for monitoring of Misato 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 Misato gene expression knockdown using RT-PCR Primer: Misato (h)-PR: sc-62619-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.