SANTA CRUZ BIOTECHNOLOGY, INC.

Twinkle siRNA (m): sc-63178

BACKGROUND

Twinkle, also known as PEO1 (progressive external ophthalmoplegia 1 protein), PEOA3, SANDO or TWINL, is a mitochondrial protein that functions as a 5'-3' nucleotide-dependent DNA helicase. Co-localized with mtDNA (mitochondrial DNA) in mitochondrial nucleoids, Twinkle is important in the metabolism and maintenance of mtDNA, playing a crucial role in the regulation of mtDNA copy numbers. Twinkle is expressed at high levels in testis, pancreas and skeletal muscle and exists as three isoforms due to alternative splicing events. Defects in the gene encoding Twinkle are the cause of two conditions: progressive external ophthalmoplegia with mitochondrial DNA deletions autosomal dominant 3 (PEOA3) and sensory ataxic neuropathy dysarthria and ophthalmoparesis (SANDO). PEOA3 is characterized by ptosis and weak muscles, while SANDO is characterized by ophthalmoparesis, dysarthria and sensory ataxic neuropathies.

REFERENCES

1. Korhonen, J.A., et al. 2003. Twinkle has 5'→3' DNA helicase activity and is specifically stimulated by mitochondrial single-stranded DNA-binding protein. J. Biol. Chem. 278: 48627-48632.
2. Wanrooij, S., et al. 2004. Twinkle and POLG defects enhance age-dependent accumulation of mutations in the control region of mtDNA. Nucleic Acids Res. 32: 3053-3064.
3. Tyynismaa, H., et al. 2004. Twinkle helicase is essential for mtDNA maintenance and regulates mtDNA copy number. Hum. Mol. Genet. 13: 3219-3227.
4. Timmons, J.A., et al. 2006. Expression profiling following local muscle inactivity in humans provides new perspective on diabetes-related genes. Genomics 87: 165-172.
5. Wanrooij, S., et al. 2007. Expression of catalytic mutants of the mtDNA helicase Twinkle and polymerase POLG causes distinct replication stalling phenotypes. Nucleic Acids Res. 35: 3238-3251.
6. Rivera, H., et al. 2007. Mild ocular myopathy associated with a novel mutation in mitochondrial twinkle helicase. Neuromuscul. Disord. 17: 677-680.
7. Baloh, R.H., et al. 2007. Familial parkinsonism and ophthalmoplegia from a mutation in the mitochondrial DNA helicase twinkle. Arch. Neurol. 64: 998-1000.
8. Sarzi, E., et al. 2007. Twinkle helicase (PEO1) gene mutation causes mitochondrial DNA depletion. Ann. Neurol. 62: 579-587.
9. Hakonen, A.H., et al. 2007. Recessive Twinkle mutations in early onset encephalopathy with mtDNA depletion. Brain 130: 3032-3040.

CHROMOSOMAL LOCATION

Genetic locus: Peo1 (mouse) mapping to 19 C3.

PROTOCOLS

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

PRODUCT

Twinkle 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Twinkle shRNA Plasmid (m): sc-63178-SH and Twinkle shRNA (m) Lentiviral Particles: sc-63178-V as alternate gene silencing products.

For independent verification of Twinkle (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-63178A, sc-63178B and sc-63178C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Twinkle siRNA (m) is recommended for the inhibition of Twinkle 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 µM in 66 µl. 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Twinkle gene expression knockdown using RT-PCR Primer: Twinkle (m)-PR: sc-63178-PR (20 µl). 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.