SANTA CRUZ BIOTECHNOLOGY, INC.

ENDOG siRNA (m): sc-144651

BACKGROUND

Endonuclease G (ENDOG), a nuclear encoded protein, localizes to the mitochondria. This sugar-nonspecific nuclease, responsible for major mitochondrial nuclease activity, preferentially cleaves single-stranded DNA(ssDNA). Synthesized as a propeptide with an amino-terminal presequence that targets the nuclease to mitochondria, ENDOG translocates to neuclei on apoptotic stimulation and act as a nuclease without sequence specificity. Both exonucleases and DNase I stimulate the ability of ENDOG to generate double-stranded DNA cleavage products at physiological ionic strengths, suggesting that these activities work in concert with ENDOG in apoptotic cells to ensure efficient DNA breakdown. In addition to deoxyribonuclease activities, ENDOG also has ribonuclease (RNase) and RNase H activities. ENDOG is capable of generating the RNA primers required by DNA polymerase γ to initiate replication of mitochondrial DNA. ENDOG exists in the mitochondrial intermembrane space, but not in the matrix where mtDNA replication occurs. This enzyme provides an important nicking function for mitochondrial DNA specifically cleaving DNA at GC tracts. Human ENDOG maps to chromosome 9q34.1.

REFERENCES

1. Cote, J., et al. 1993. Primers for mitochondrial DNA replication generated by endonuclease G. Science 5122: 765-769.
2. Tiranti V., et al. 1995. Chromosomal localization of mitochondrial transcription factor A (TCF6), single-stranded DNA-binding protein (SSBP), and endonuclease G (ENDOG), three human housekeeping genes involved in mitochondrial biogenesis. Genomics 2: 559-564.
3. Widlak, P., et al. 2001. Action of recombinant human apoptotic endonuclease G on naked DNA and chromatin substrates: cooperation with exonuclease and DNase I. J. Biol. Chem. 51: 48404-48409.
4. Ikeda, S., et al. 2002. Mitochondrial factors modulate the activity of endonuclease G, the major nuclease of Mammalian mitochondria. J. Biochem. Mol. Biol. Biophys. 1: 17-21.
5. Ohsato, T., et al. 2002. Mammalian mitochondrial endonuclease G. Digestion of R-loops and localization in intermembrane space. Eur. J. Biochem. 23: 5765-5770.
6. LocusLink Report (LocusID: 2021). http://www.ncbi.nlm.nih.gov/LocusLink/

CHROMOSOMAL LOCATION

Genetic locus: Endog (mouse) mapping to 2 B.

PRODUCT

ENDOG siRNA (m) is a target-specific 19-25 nt siRNA 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 ENDOG shRNA Plasmid (m): sc-144651-SH and ENDOG shRNA (m) Lentiviral Particles: sc-144651-V as alternate gene silencing products.

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 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

ENDOG siRNA (m) is recommended for the inhibition of ENDOG 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.

GENE EXPRESSION MONITORING

ENDOG (B-2): sc-365359 is recommended as a control antibody for monitoring of ENDOG 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ENDOG gene expression knockdown using RT-PCR Primer: ENDOG (m)-PR: sc-144651-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.