

DNA pol α siRNA (h): sc-37771

BACKGROUND

DNA replication, recombination and repair, all of which are necessary for genomic stability, require the presence of exonucleases. In DNA replication, these enzymes are involved in the processing of Okazaki fragments, whereas in DNA repair, they function to excise damaged DNA fragments and correct recombinational mismatches. These exonucleases include the family of DNA polymerases. DNA pol α , β , δ and ϵ are involved in DNA replication and repair. DNA pol δ and DNA pol ϵ are multisubunit enzymes, with DNA pol δ consisting of two subunits: p125, which interacts with the sliding DNA clamp protein PCNA; and p50. The nuclear-encoded DNA pol γ is the only DNA polymerase required for the replication of the mitochondrial DNA. DNA pol ζ is ubiquitously expressed in various tissues and mediates the cellular mechanism of damage-induced mutagenesis. DNA pol θ is a DNA polymerase-helicase that binds ATP and is involved in the repair of interstrand crosslinks.

REFERENCES

1. Bambara, R.A., et al. 1991. Properties of DNA polymerases δ and ϵ , and their roles in eukaryotic DNA replication. *Biochim. Biophys. Acta* 1088: 11-24.
2. Li, J.J., et al. 1992. DNA replication. Eukaryotic initiation rites. *Nature* 357: 114-115.
3. Ropp, P.A., et al. 1996. Cloning and characterization of the human mitochondrial DNA polymerase, DNA polymerase γ . *Genomics* 36: 449-458.
4. Kolodner, R.D., et al. 1999. Eukaryotic DNA mismatch repair. *Curr. Opin. Genet. Dev.* 9: 89-96.
5. Wood, R.D. 1999. DNA repair: Variants on a theme. *Nature* 399: 639-640.
6. Diede, S.J., et al. 1999. Telomerase-mediated telomere addition *in vivo* requires DNA primase and DNA polymerases α and δ . *Cell* 99: 723-733.
7. Lin, W., et al. 1999. A full-length cDNA of hREV3 is predicted to encode DNA polymerase ζ for damage-induced mutagenesis in humans. *Mutat. Res.* 433: 89-98.
8. Sharief, F.S., et al. 1999. Cloning and chromosomal mapping of the human DNA polymerase theta (POLQ), the eighth human DNA polymerase. *Genomics* 59: 90-96.

CHROMOSOMAL LOCATION

Genetic locus: POLA (human) mapping to Xp22.11.

PRODUCT

DNA pol α 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 DNA pol α shRNA Plasmid (h): sc-37771-SH and DNA pol α shRNA (h) Lentiviral Particles: sc-37771-V as alternate gene silencing products.

For independent verification of DNA pol α (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-37771A, sc-37771B and sc-37771C.

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

DNA pol α siRNA (h) is recommended for the inhibition of DNA pol α 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

DNA pol α (D-7): sc-137021 is recommended as a control antibody for monitoring of DNA pol α 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 DNA pol α gene expression knockdown using RT-PCR Primer: DNA pol α (h)-PR: sc-37771-PR (20 μ l, 534 bp). 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.

PROTOCOLS

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