



DNA pol ι siRNA (m): sc-37786

BACKGROUND

DNA polymerase activity is essential for replication, repair, recombination and mutagenesis. DNA polymerases can often bypass DNA lesions that block DNA replication, thereby allowing the replication of damaged DNA. One such DNA polymerase is the distributive enzyme DNA pol ι (pol iota), which is encoded by the POLI gene. POLI is located on human chromosome 18q21.2, a region often implicated in the etiology of many human cancers. At thymine templates, DNA Pol ι is highly error-prone when replicating undamaged DNA in that it favors the misincorporation of guanine over the correct nucleotide, adenosine. DNA Pol ι also promotes the replication of damaged DNA by misincorporating deoxynucleotides opposite DNA lesions. DNA Pol ι acts sequentially with DNA Pol ζ , which is essential for damage-induced mutagenesis, to complete the DNA lesion bypass. Therefore, replication involving DNA Pol ι is likely to be highly mutagenic.

REFERENCES

1. Johnson, R.E., Washington, M.T., Haracska, L., Prakash, S. and Prakash, L. 2000. Eukaryotic polymerase ι and ζ act sequentially to bypass DNA lesions. *Nature* 406: 1015-1019.
2. Tissier, A., Frank, E.G., McDonald, J.P., Iwai, S., Hanaoka, F. and Woodgate, R. 2000. Misinsertion and bypass of thymine-thymine dimers by human DNA polymerase ι . *EMBO J.* 19: 5259-5266.
3. Tissier, A., McDonald, J.P., Frank, E.G. and Woodgate, R. 2000. Novel human and mouse homologs of *Saccharomyces cerevisiae* DNA polymerase η . *Genomics* 60: 20-30.
4. Tissier, A., McDonald, J.P., Frank, E.G. and Woodgate, R. 2000. pol ι , a remarkably error-prone human DNA polymerase. *Genes Dev.* 14: 1642-1650.
5. Zhang, Y., Yuan, F., Wu, X. and Wang, Z. 2000. Preferential incorporation of G opposite template T by the low-fidelity human DNA polymerase ι . *Mol. Cell. Biol.* 20: 7099-7108.

CHROMOSOMAL LOCATION

Genetic locus: Poli (mouse) mapping to 18 E2.

PRODUCT

DNA pol ι siRNA (m) is a pool of 3 target-specific 20-25 nt siRNAs designed to knock down gene expression. Each vial contains 3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections.

For independent verification of DNA pol ι (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-37786A, sc-37786B and sc-37786C.

PROTOCOLS

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

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

DNA pol ι siRNA (m) is recommended for the inhibition of DNA pol ι 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 μ M in 60 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 ι (M-14): sc-14255 is recommended as a control antibody for monitoring of DNA pol ι 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor DNA pol ι gene expression knockdown using RT-PCR Primer: DNA pol ι (m)-PR: sc-37786-PR (20 μ l, 526 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.