SANTA CRUZ BIOTECHNOLOGY, INC.

DNA Ligase I siRNA (m): sc-35199

BACKGROUND

DNA Ligase I maintains the major DNA Ligase activity in proliferating cells by joining Okazaki fragments during lagging strand DNA replication. Human DNA Ligase I also has an essential role in DNA repair pathways, where it catalyzes the formation of phosphodiester bonds between adjacent 5' phosphoryl and 3' hydroxy termini at single breaks in duplex DNA molecules. In addition, DNA Ligase I plays a role in sealing nicks during excision repair. Similar to other DNA Ligases, DNA Ligase I is built around a common catalytic core. Increased levels of DNA Ligase I are found in human tumors, as compared to benign tissues, as well as in peripheral blood lymphocytes. Furthermore, DNA Ligase I antisense ODNs may decrease tumor cell proliferation, suggesting a potential role for DNA Ligase I as an anti-cancer agent. DNA Ligase I activity is altered in the chromosomal breakage deficit Bloom's syndrome (BS). Individuals with BS either have decreased levels of abnormally thermolabile DNA Ligase I or possess a dimeric form of this enzyme.

REFERENCES

1. Barnes, D.E., et al. 1990. Human DNA Ligase I cDNA: cloning and functional expression in *Saccharomyces cerevisiae*. Proc. Natl. Acad. Sci. USA 87: 6679-6683.
2. Petrini, J.H., et al. 1991. A wild-type DNA Ligase I gene is expressed in Bloom's syndrome cells. Proc. Natl. Acad. Sci. USA 88: 7615-7619.
3. Barnes, D.E., et al. 1992. Mutations in the DNA Ligase I gene of an individual with immunodeficiencies and cellular hypersensitivity to DNA-damaging agents. Cell 69: 495-503.
4. Timson, D.J., et al. 2000. DNA Ligases in the repair and replication of DNA. Mutat. Res. 460: 301-318.
5. Sun, D., et al. 2001. Elevated expression of DNA Ligase I in human cancers. Clin. Cancer Res. 7: 4143-4148.
6. Tom, S., et al. 2001. DNA Ligase I and proliferating cell nuclear antigen form a functional complex. J. Biol. Chem. 276: 24817-24825.

CHROMOSOMAL LOCATION

Genetic locus: Lig1 (mouse) mapping to 7 A1.

PRODUCT

DNA Ligase I 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 DNA Ligase I shRNA Plasmid (m): sc-35199-SH and DNA Ligase I shRNA (m) Lentiviral Particles: sc-35199-V as alternate gene silencing products.

For independent verification of DNA Ligase I (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-35199A, sc-35199B and sc-35199C.

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 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

DNA Ligase I siRNA (m) is recommended for the inhibition of DNA Ligase I 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

DNA Ligase I (C-5): sc-271678 is recommended as a control antibody for monitoring of DNA Ligase I 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Ligase I gene expression knockdown using RT-PCR Primer: DNA Ligase I (m)-PR: sc-35199-PR (20 µl). 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.