SANTA CRUZ BIOTECHNOLOGY, INC.

Topo I siRNA (h): sc-36694

BACKGROUND

DNA topoisomerase I and II (Topo I and Topo II) are nuclear enzymes that regulate the topological structure of DNA in eukaryotic cells by transiently breaking and rejoining DNA strands. Eukaryotic topoisomerases are capable of relaxing both positive and negative supercoils, whereas prokaryotic topoisomerases relax only negative supercoils. DNA topoisomerases play a role in DNA replication, recombination, and transcription and have been identified as targets of numerous anticancer drugs. Topo I, a ubiquitously expressed, soluble enzyme, acts by introducing a transient break in one strand of DNA, while Topo II acts by making a transient double-strand break. Topo II is encoded by two different genes to generate two distinct isoforms that are designated Topo IIα and Topo IIβ. Topo IIβ, and Topo IIα are largely homologous at their N-terminal three quarters, however, the C-terminal segments are considerably divergent, suggesting that these regions may mediate different cellular functions and account for the observed differential tissue expression patterns of the two isoforms.

REFERENCES

1. D'Arpa, P., et al. 1988. cDNA cloning of human DNA topoisomerase I: catalytic activity of a 67.7 kDa carboxyl-terminal fragment. Proc. Natl. Acad. Sci. USA 85: 2543-2547.
2. Chung, T.D., et al. 1989. Characterization and immunological identification of cDNA clones encoding two human DNA topoisomerase II isozymes. Proc. Natl. Acad. Sci. USA 86: 9431-9435.
3. Austin, C.A., et al. 1990. Isolation and characterization of a human cDNA clone encoding a novel DNA topoisomerase II homologue from HeLa cells. FEBS Lett. 266: 115-117.

CHROMOSOMAL LOCATION

Genetic locus: TOP1 (human) mapping to 20q12.

PRODUCT

Topo I 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Topo I shRNA Plasmid (h): sc-36694-SH and Topo I shRNA (h) Lentiviral Particles: sc-36694-V as alternate gene silencing products.

For independent verification of Topo I (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-36694A, sc-36694B and sc-36694C.

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

Topo I siRNA (h) is recommended for the inhibition of Topo I 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 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

Topo I (C-21): sc-32736 is recommended as a control antibody for monitoring of Topo 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Topo I gene expression knockdown using RT-PCR Primer: Topo I (h)-PR: sc-36694-PR (20 µl, 422 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ganguly, A., et al. 2007. Betulinic acid, a catalytic inhibitor of topoisomerase I, inhibits reactive oxygen species-mediated apoptotic topoisomerase I-DNA cleavable complex formation in prostate cancer cells but does not affect the process of cell death. Cancer Res. 67: 11848-11858.
2. Wang, P., et al. 2009. Topoisomerase I and RecQL1 function in Epstein-Barr virus lytic reactivation. J. Virol. 83: 8090-8098.
3. Velichko, A.K., et al. 2015. Mechanism of heat stress-induced cellular senescence elucidates the exclusive vulnerability of early S-phase cells to mild genotoxic stress. Nucleic Acids Res. 43: 6309-6320.
4. Inoue, N., et al. 2021. The benzylisoquinoline alkaloids, berberine and coptisine, act against camptothecin-resistant topoisomerase I mutants. Sci. Rep. 11: 7718.
5. Nishida, M., et al. 2022. Mechanism of action of non-camptothecin inhibitor Genz-644282 in topoisomerase I inhibition. Commun. Biol. 5: 982.

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.