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Product Information

Topoisomerase I, human
recombinant, expressed in baculovirus

Catalog Number **T9069**
Storage Temperature –70 °C

EC 5.99.1.2

Product Description

Topoisomerase I plays a major role in critical cellular processes by catalyzing the breakage and religation of phosphodiester bonds in a single strand of DNA. This results in the removal of supercoils (either positive or negative superhelical turns).[1,2] Topoisomerase I is important in vital processes including replication, transcription, and recombination.[1,3]

The enzyme is used to study DNA structure and topology, such as the effects of supercoiling on transcription *in vitro,* chromatin reconstitution *in vitro*, and the degree of supercoiling of DNA. It can also be used to assay mutant plasmids, which differ in length by only one base-pair and to increase restriction endonuclease digestion of resistant DNA substrates by unwinding the DNA coils to expose restriction sites.

Molecular mass: 100 kDa (SDS-PAGE)

Topoisomerase I is expressed in baculovirus and purified to homogeneity. It is supplied in a solution of 20 mM NaH_2PO_4, pH 7.4, 300 mM NaCl, 50 µg/mL BSA, 50% glycerol, and 25–100 mM imidazole.

This product is free of detectable contaminating proteins including topoisomerase II and nuclease. The enzyme is not certified for use in Western blotting applications.

Activity: ≥2 units/µL

Unit definition: One unit will relax 0.25 µg of supercoiled plasmid DNA in 30 minutes at pH 7.9 at 37 °C.

Assay Buffer: 10 mM Tris-HCl, pH 7.9, with 150 mM NaCl, 0.1% BSA, 0.1 mM spermidine, and 5% glycerol.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

Store the product at –70 °C. It is recommended the product be aliquoted after the first thaw to avoid repeated freezing and thawing, which may result in spontaneous degradation of the 100 kDa protein to 60–80 kDa bands.

In general the enzyme should **not** be diluted before freezing at –70 °C as it loses stability when diluted.

References

1. Pommier, Y. *et al*., Mechanism of action of eukaryotic DNA topoisomerase I and drugs targeted to the enzyme. Biochim. Biophys. Acta, **1400**, 83-105 (1998).
2. Stewart, L. and Champoux, J.J., Assaying DNA topoisomerase I relaxation activity. Methods Mol. Biol., **95**, 1-11 (2001).
3. Lisby, M. *et al*., Residues within the N-terminal domain of human topoisomerase I play a direct role in relaxation. J. Biol. Chem., **276**, 20220-20227 (2001).

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