

# *S.pneumoniae* Topoisomerase IV



## Product Description (Product Numbers SPT4001 and SPT4002)

*S.pneumoniae* Topoisomerase IV is prepared by overexpressing the parC and parE subunits in *E. coli* and purifying them by in-house methods adapted from Peng and Mariani, 1999. It is supplied as a heterotetramer complex. The enzyme is supplied at a minimum concentration of 5 U/μl in Dilution Buffer. Store at -80 °C. (Stable for 6 months undiluted.) It is recommended that the enzyme is aliquoted to avoid repeated freeze-thaw cycles.

**For *in vitro* laboratory research use only.**

### Dilution Buffer

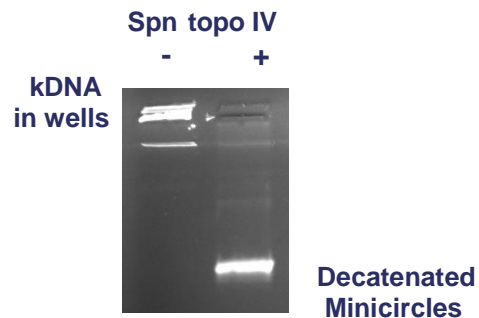
40 mM HEPES.KOH (pH 7.6)  
100 mM potassium glutamate  
1 mM DTT  
1 mM EDTA  
40 % (v/v) glycerol

### Assay Buffer (supplied as 5X stock)

40 mM Tris-HCl (pH 7.5),  
6 mM MgCl<sub>2</sub>,  
10 mM NaCl,  
10 mM DTT,  
200 mM potassium glutamate,  
1 mM ATP,  
0.05 mg/ml BSA

### Decatenation Assay

1 U of *S.pneumoniae* topoisomerase IV will decatenate 0.2 μg of kDNA when incubated in Assay Buffer in a total reaction volume of 30 μl at 37 °C for 30 minutes.  
Gels are run in the absence of ethidium bromide or chloroquine.



### Quality Control

Purity: The parC and parE subunits are purified to > 95 % purity as judged by SDS-polyacrylamide gel electrophoresis.

### Reference

Peng, H. and Mariani, K.J. (1999). Overexpression and purification of bacterial topoisomerase IV, in *DNA Topoisomerase Protocols* Vol. I (Bjornsti, M-A., and Osheroff, N. eds.), Humana Press, Totowa, N.Jersey pp.163-169