

M. tuberculosis Topoisomerase I Assay Kit



Product Description (Product Numbers MTTR01 and MTTR02)

M. tuberculosis topoisomerase I is purified after heterologous expression in *E. coli*.

The enzyme is supplied at a minimum concentration of 10 U/ μ l in Dilution Buffer. However, we recommend that the enzyme is titrated into the assay to ascertain the minimum volume of enzyme required per assay to achieve full relaxation.

Store at -80 °C. It is recommended that the enzyme is aliquoted to avoid repeated freeze-thaw cycles.

For *in vitro* laboratory research use only.

Dilution Buffer

50 mM Tris.HCl (pH 7.6)
1 mM DTT
1 mM EDTA
50 % (v/v) glycerol
50 mM NaCl

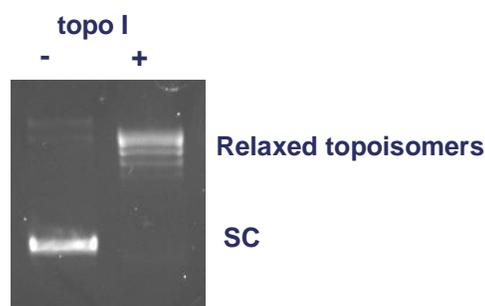
Assay Buffer (supplied as 5X stock)

40 mM Tris.HCl (pH7.6)
20 mM NaCl
1 mM EDTA
5 mM MgCl₂
50 μ g/ml albumin

Relaxation Assay

1 U of topoisomerase I will relax 0.5 μ g of supercoiled pBR322 when incubated in Assay Buffer in a total reaction volume of 30 μ l at 37 °C for 30 minutes.

Gels are run in the absence of ethidium bromide or chloroquine.



Quality Control

1) Purity: *M. tuberculosis* topoisomerase I is purified to > 95 % purity as judged by SDS-polyacrylamide gel electrophoresis. 2) pBR322 were also incubated for 4hrs in assay buffer (+ 10 mM MgCl₂) at 37 °C. These tests were negative for the formation of linear products, showing the absence of nuclease contamination.