# M. tuberculosis Gyrase Assay Kit for cell extracts



#### **Product Description** (MTE001 and MTE00E)

The kit is designed for the detection of M.tuberculosis gyrase activity in cell extracts. Store at - 20 °C. Includes relaxed pBR322 DNA substrate, assay and dilution buffers and supercoiled pBR322 DNA marker.

For in vitro laboratory research use only.

#### **Dilution Buffer (1X)**

50 mM Tris.HCl (pH 7.9) 5 mM DTT 30 % (w/v) glycerol

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

50 mM HEPES. KOH (pH 7.9)

6 mM magnesium acetate

4 mM DTT

1 mM ATP

100 mM potassium glutamate

2 mM spermidine

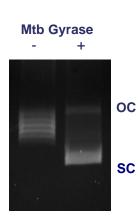
0.05 mg/ml albumin

### **Supercoiling Assay**

1 U of Mtb gyrase/cell extract is incubated with 0.5  $\mu$ g of relaxed pBR322 in a reaction volume of 30  $\mu$ l at 37°C for 30 minutes in Assay Buffer.

Gels are run in the absence of Ethidium Bromide or Chloroquine.

## Relaxed Topoisomers



#### **Quality Control**

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

3) No activity was detectable when the single subunits were assayed alone