

RELAXED pBR322



Product Description (Product Numbers R5001, R2502, R5003 and R1004)

Supercoiled plasmid pBR322 DNA is produced by the large-scale alkaline-lysis method (Sambrook et al., 1989) from a high-copy number derivative of pBR322 (Boros et al., 1984) and is relaxed using chicken erythrocyte topoisomerase I (Trask and Muller, 1983).

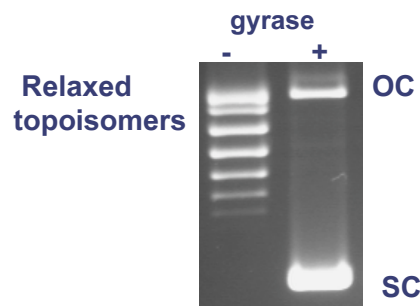
It is shipped on dry ice at a concentration of 1mg/ml in TE.
Store at 4°C. For in-vitro laboratory research use only.

TE Storage Buffer

10mM Tris-HCL (pH 7.5)
1mM EDTA

Supercoiling Assay

0.5 µg of relaxed pBR322 when incubated with 1 U of DNA gyrase in a reaction volume of 30 µl at 37°C for 30 minutes in Incubation Buffer, is completely converted to the supercoiled form.



Quality Control

Purity was determined spectroscopically using the ratio between the OD₂₆₀ and OD₂₈₀ readings.

Incubation under gyrase assay conditions at 37°C for 30 minutes did not result in any detectable conversion of the DNA to either open circular or linear forms when assessed by agarose gel electrophoresis.

References

- Boros, I., Pósfai, G. & Venetianer, P. (1984). High-copy –number derivatives of the plasmid cloning vector pBR322. *Gene* 30, 257-260
- Sambrook, J., Fritsch, E.F. & Maniatis, T (1989). *Molecular cloning: a laboratory manual*. Cold Spring Harbor Press. Cold Spring Harbor.
- Trask, D. K. & Muller, M. T. (1983) Biochemical characterization of topoisomerase I purified from avian erythrocytes. *Nucleic Acids Res.* 11, 2779-2800