S. aureus DNA Gyrase Cleavage Assay Kit

Product Description (Product Numbers SAGC001 and SAGC002)

S. aureus DNA gyrase was purified after heterologous expression in *E. coli* and is supplied as an A_2B_2 complex. The enzyme is supplied at a concentration of 4.0 µM in Dilution Buffer and is suitable for cleavage assays. Cleavage activity is 2 U/µl.
Up to 50 % cleavage can be obtained with 0.5 µl in the presence of 10 µM CFX in a typical 30 µl reaction (see typical titration below).
Store at -80°C.
For *in vitro* laboratory research use only.

Dilution Buffer

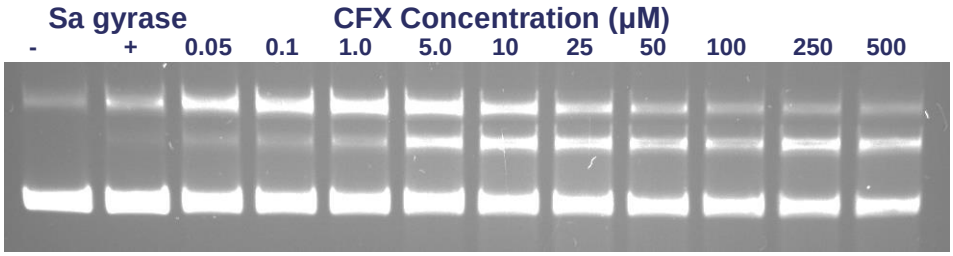
50 mM Tris.HCl (pH 7.5)
1 mM DTT
1 mM EDTA
40 % (w/v) glycerol

Assay Buffer (supplied as 5X stock)

40 mM HEPES. KOH (pH 7.6)
10 mM magnesium acetate
10 mM DTT
100 mM potassium glutamate
0.05 mg/ml albumin

Cleavage Assay

S. aureus DNA gyrase is incubated with 0.5 µg of supercoiled pBR322 in a reaction volume of 30 µl at 37 °C for 1 hour in Assay Buffer in the presence of CFX.
0.2 % SDS and 0.1 mg/ml Proteinase K are added before a further incubation at 37 °C for 30 minutes.



Quality Control

1) Purity: *S. aureus* DNA 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 $MgCl_2$ 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.