

Escherichia coli Gyrase wildtype B Subunit



Product Description (Product Numbers SUBB001, SUBB005, SUBB100)

DNA gyrase B subunit is prepared from the over producing strain JMtacA (Hallett, *et al.*, 1990). Supercoiling activity is 60-100U/ul when tested in combination with the GyrA subunit.

Store at -80°C.

For *in vitro* laboratory research use only.

Dilution Buffer

50 mM Tris.HCl (pH 7.5)
100 mM KCl
2 mM dithiothreitol
1 mM EDTA
50 % (w/v) glycerol

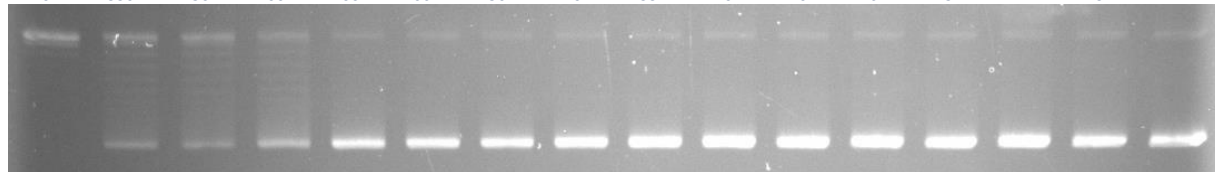
Assay Buffer (supplied as 5X stock)

35 mM Tris.HCl (pH 7.5)
24 mM KCl
4 mM MgCl₂
2 mM dithiothreitol
1.8 mM spermidine
1 mM ATP
6.5 % (w/v) glycerol
0.1 mg/ml albumin

Supercoiling Assay

E.coli gyrase is incubated with 0.5 µg of relaxed pBR322 in a reaction volume of 30 µl at 37°C for 30 minutes in Assay Buffer. Gels are run in the absence of Ethidium Bromide or Chloroquine. Supercoiling activity is 60-100U/ µl. Protein concentration is 1mg/ml

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	Tube
0	330	250	200	100	66	50	40	33	20	10	6	5	4	3	2	U/ul



1 U of *E.coli* gyrase is defined as the amount of enzyme required to relax 0.5 µg of pBR322 in a reaction volume of 30 µl at 37°C for 30 minutes in Assay Buffer.

Reference

Hallett, P., Grimshaw, A.J., Wigley, D.B. and Maxwell, A. (1990) Cloning of the DNA gyrase genes under *tac* promoter control: overproduction of the gyrase A and B proteins. *Gene* 93: 139-142

***Escherichia coli* Gyrase wildtype A Subunit**



Product Description (Product Numbers SUBA001, SUBA005, SUBA100)

DNA gyrase A subunit is prepared from the over producing strain JMtacA (Hallett, *et al.*, 1990). Supercoiling activity is 60-100U/ul when tested in combination with the GyrB subunit.

Store at -80°C.

For *in vitro* laboratory research use only.

Dilution Buffer

50 mM Tris.HCl (pH 7.5)
100 mM KCl
2 mM dithiothreitol
1 mM EDTA
50 % (w/v) glycerol

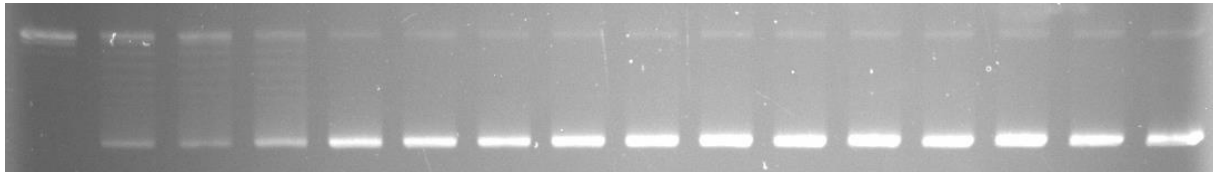
Assay Buffer (supplied as 5X stock)

35 mM Tris.HCl (pH 7.5)
24 mM KCl
4 mM MgCl₂
2 mM dithiothreitol
1.8 mM spermidine
1 mM ATP
6.5 % (w/v) glycerol
0.1 mg/ml albumin

Supercoiling Assay

E.coli gyrase is incubated with 0.5 µg of relaxed pBR322 in a reaction volume of 30 µl at 37°C for 30 minutes in Assay Buffer. Gels are run in the absence of Ethidium Bromide or Chloroquine. Supercoiling activity is 60-100U/ µl. Protein concentration is 1mg/ml

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	Tube
0	330	250	200	100	66	50	40	33	20	10	6	5	4	3	2	U/ul



1 U of *E.coli* gyrase is defined as the amount of enzyme required to relax 0.5 µg of pBR322 in a reaction volume of 30 µl at 37°C for 30 minutes in Assay Buffer.

Reference

Hallett, P., Grimshaw, A.J., Wigley, D.B. and Maxwell, A. (1990) Cloning of the DNA gyrase genes under *tac* promoter control: overproduction of the gyrase A and B proteins. *Gene* 93: 139-142