T4 DNA Ligase

Code No. LGA-111

Lot No. ******

Storage Store at −20℃

Size 400units

Source : *Escherichia coli* lysogenic for NM989

Reaction : ATP + $(dNMP)_n$ + $(dNMP)_m$ → AMP + PPi + $(dNMP)_{n+m}$

Concentration : ＊＊＊ units/μl

Unit Definition : One unit is the amount of enzyme activity that catalyzes the conversion of 1 nmole of ^{32}PPi into a Norit absorbable form in 20 minutes at 37℃.

Assay Condition :

66	mM	Tris-HCl(pH7.6)
6.6	mM	$MgCl_2$
10	mM	DTT
66	μM	ATP
3.3	μM	(^{32}P) $Na_4P_2O_7$

Storage Buffer :

20	mM	Tris-HCl(pH7.6)
1	mM	EDTA
5	mM	DTT
60	mM	KCl
50	%	Glycerol

10×Ligation Buffer :

660	mM	Tris-HC1(pH7.6)
66	mM	$MgCl_2$
100	mM	DTT
660	μM	ATP＊

＊ ATP is not included in the attached buffer .

Contaminant Assay

1. Nonspecific Endonuclease : When 10 units of enzyme were incubated with 1μg of *Hin*dⅢ digest of λ-DNA for 20 hours at 16℃ in 50 μl reaction volume , no degradation of the DNA frgaments is observed after agarose gel electrophoresis .

2. Nonspecific Exonuclease : 30 units of enzyme, when incubated with 1μg of *E. coli* ^{3}H-DNA for 4 hours at 16℃ in 50 μl reaction volume, will release less than 0.01 % acid soluble counts.

3. 5 '-Exonuclease and 5 '-Phosphatase : 30 units of enzyme, when incubated with 5′-^{32}P-termini labeled *Hin*dⅢ digest of λ-DNA for 4 hours at 16℃ in 50 μl reaction volume, will release less than 0.01 % acid soluble counts.

4. Nicking Activity : When 10 units of enzyme were incubated with 1μg of ΦX174 DNA(RFI) for 20 hours at 16℃ in 50 μl reaction volume , no relaxing of the supercoil

structure is observed after agarose gel electrophoresis .