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ISO9001

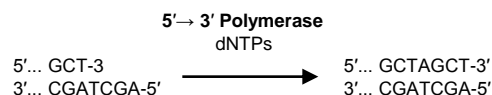
## DNA Polymerase I Large (Klenow-exo) Fragment

| Product   | Quantity | Cat. No. | Remarks         |
|---|----------|----------|-----------------|
| DNA Polymerase I Large<br>(Klenow-exo) Fragment | 200 unit | EBT-3000 | 5 unit/ $\mu$ l |

### Description

DNA Polymerase I Large (Klenow -exo) Fragment is a DNA-dependent DNA polymerase that lacks both the 5'→3' and 3'→5' exonuclease activity of intact *E. coli* DNA Polymerase I. DNA Polymerase I Large (Klenow -exo) Fragment is used for random priming and chain displacement amplification. But it is not recommended in blunt formation for blunt-ended ligation.

DNA Polymerase I Large (Klenow -exo) Fragment is purified from a recombinant *E.coli* strain.



### Concentration & Storage Condition

5 unit/  $\mu$ l. Store at -20°C.

### Storage Buffer

50 mM Tris-HCl, pH 7.5, 1 mM DTT, 0.1 mM EDTA, 50% (v/v) glycerol.

### 10x Reaction Buffer

500 mM Tris-HCl, pH 7.2, 100 mM MgSO<sub>4</sub>, 1 mM DTT.

### Unit Definition

One unit is defined as the amount of enzyme that incorporates 10nmole of total dNTP into TCA-insoluble material in 30 min at 37°C. The reaction conditions are: 67 mM potassium phosphate, pH 7.5, 6.7 mM MgCl<sub>2</sub>, 1 mM DTT, 50  $\mu$ g/ml activated calf thymus DNA and 33 mM each of dATP, dCTP, dGTP and dTTP (a mix of unlabeled and [<sup>3</sup>H]dTTP).

### QC Tests

Activity, exo and endonuclease activity test, SDS-PAGE purity, performance tests.

### Usage Information

#### Fill-In of 5'-Overhang to form Blunt ends

- Add the following components to the microcentrifuge tube :
 

|  |             |
|--|-------------|
| DNA (1–5 $\mu$ g digested DNA containing 5'-overhangs) | x $\mu$ l   |
| 10x Reaction Buffer                                    | 2 $\mu$ l   |
| 1mM dNTP mixture (0.25mM each)                         | 5 $\mu$ l   |
| DNA Polymerase I (2.5 units)                           | 0.5 $\mu$ l |
| Nuclease-Free Water to final volume                    | 20 $\mu$ l  |
- Incubate at 37°C for 1 hour.
- Heat at 75°C for 10 minutes to inactivate the enzyme.