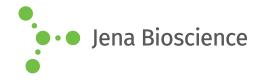
# **DATA SHEET**





# Klenow Fragment

Large Fragment of DNA Polymerase I recombinant, *E. coli* 

Cat. No.	Amount
EN-148S	300 Units
EN-148L	5 x 300 Units

**Unit Definition:** One unit is defined as the amount of enzyme required to convert 10 nmoles of dNTPs to an acid insoluble form in 30 minutes at 37 °C.

For general laboratory use.

Shipping: shipped on gel packs
Storage Conditions: store at -20 °C

Additional Storage Conditions: avoid freeze/thaw cycles

Shelf Life: 12 months

Form: liquid (Supplied in 100 mM KPO<sub>4</sub> pH 6.5, 1 mM DTT and 50 %

[v/v] glycerol)

Concentration: 5 units/µl

# **Applications:**

- Fill-in of 5' overhangs to form blunt ends
- · Removal of 3' overhangs to form blunt ends

### **Description:**

Klenow Fragment is the large fragment of DNA Polymerase I that retains its  $5' \rightarrow 3'$  polymerase,  $3' \rightarrow 5'$  exonuclease and strand displacement activities. The enzyme lacks the  $5' \rightarrow 3'$  exonuclease activity of intact DNA polymerase I. Klenow retains the polymerization fidelity of the holoenzyme without degrading 5' termini.

#### **Reaction conditions:**

- Dissolve 0.1 4  $\mu g$  of digested DNA in 1x Reaction Buffer supplemented with 40  $\mu M$  each dNTP
- Add 1 unit Klenow Fragment per μg DNA

 $\underline{\text{Incubate}}$  for 15 min. at 25 °C

Stop reaction by alternativly

- add EDTA to 10 mM final concentration
- Heat inactivation: 20 min. at 75 °C

## 10x Reaction Buffer:

500 mM Tris-HCl pH 7.6 at 25 °C, 50 mM MgCl $_2$  and 10 mM DTT.

#### Note

Excessive amounts of enzyme or longer reaction times may result in recessed ends due to the  $3' \rightarrow 5'$  exonuclease activity of the enzyme.

## **Quality Control:**

The enzyme is greater than 98 % pure as indicated by SDS-polyacrylamide gel electrophoresis and contains no detected endonuclease activity. Incubation of 10 units of Klenow with supercoiled plasmid DNA produced no nicked molecules after 20 hours at 37 °C as determined by agarose gel electrophoresis analysis.

