

Intact Genomics® **DNA Polymerase I,  
Large (Klenow) Fragment**

|                      |            |             |
|----------------------|------------|-------------|
| <b>Catalog #</b>     | 3261       | 3262        |
| <b>Package Size</b>  | 200 units  | 1,000 units |
| <b>Volume</b>        | 40 µl      | 200 µl      |
| <b>Concentration</b> | 5 units/µl |             |

### Description

Large Fragment of DNA Polymerase I (Klenow) is a product of *E. coli* DNA Polymerase I which lacks the N-terminal 324 amino acids. This enzyme lacks the 5'→3' exonuclease activity of intact DNA Polymerase I, but does exhibit the 5'→3' DNA polymerase and 3'→5' exonuclease activities

### Protein Purity

The physical purity of this enzyme is ≥99% as assessed by SDS-PAGE with Coomassie® blue staining (see figure below).

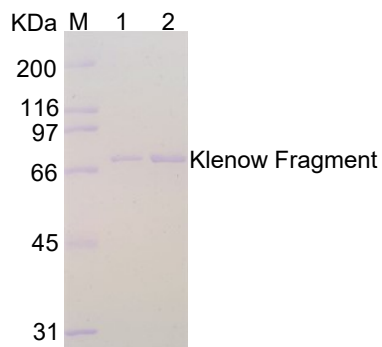


Figure: Lane M. Protein Marker  
Lane 1. 0.5 µg of Klenow Fragment  
Lane 2. 1.0 µg of Klenow Fragment

### Product Source

*E. coli* cells carrying *E. coli* polA gene without its N-terminal exonuclease domain.

### Applications

- 3'-overhangs removal to form blunt ends (1).
- 5'-overhangs fill-in to form blunt ends (1).
- DNA library preparation for Next-generation sequencing (2).
- 3'-end-labeling of DNA fragments using  $\alpha$ -<sup>32</sup>P deoxynucleotides.
- Second strand cDNA synthesis.
- Single-stranded DNA probes generation.

### Product Includes

- 1) DNA Polymerase I, Large (Klenow) Fragment
- 2) 10x Klenow Reaction Buffer

### Storage Temperature

-20 °C

### Storage Buffer

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

### 10x Klenow Reaction Buffer

500 mM Tris-HCl  
100 mM MgCl<sub>2</sub>  
50 mM dithiothreitol (DTT)  
pH 7.5 @ 25 °C.

### Inactivation

Inactivated by heating at 75 °C for 20 min.

### Quality Control Assays

DNA Polymerase I, Klenow Fragment is free from detectable endonuclease and RNase activities.

### References

1. Sambrook, J., Fritsch, E.F. and Maniatis, T. (1989). *Molecular Cloning: A Laboratory Manual*. (2nd ed.), 5.40-5.43. Cold Spring Harbor: Cold Spring Harbor Laboratory Press.
2. Sanger, F. et al. (1977). *Proc. Natl. Acad. Sci. USA*. 74, 5463-5467.