

Phi29 DNA Polymerase

Catalog #PHI-BE101

Product Component	Sizes
Phi29 DNA Polymerase (10U/μL)	200U, 500U, 5000U
10X Phi29 DNA Polymerase Reaction Buffer	200μL, 500μL, 5mL

Storage/Transportation Condition Store at -20°C ± 5°C for up to 24 months. Avoid repeated freeze/thaw cycles. Transport on dry ice.

Form Liquid

Source *E. coli* strain carrying the phi29 DNA polymerase gene cloned from bacteriophage phi29.

Storage Buffer 10 mM Tris, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.5% Tween 20, 50% Glycerol, pH 7.4

10X phi29 DNA Polymerase Reaction Buffer 500 mM Tris-HCl, 100 mM MgCl₂, 100 mM (NH₄)₂SO₄, 40 mM DTT, PH 7.5

Concentration 10U/μL

Unit Definition One unit is defined as the amount of enzyme required to incorporate 0.5 pmol of dNTPs into acid-insoluble substances within 10 min in a total reaction volume of 50 μL at 30°C.

Product Description

Phi29 DNA polymerase is a DNA polymerase cloned from *Bacillus subtilis* phage phi29 with strong strand displacement activity and high processivity (up to more than 70 kbp). The enzyme has inherent 3'-5' exonuclease activity and higher fidelity than most Taq enzymes. It is an enzyme used for isothermal amplification *in vitro*.

Applications

- Multiple strand displacement amplification (MDA)
- Whole genome amplification (WGA)
- Rolling circle amplification
- Protein-primed replication
- Cell-free cloning

Recommended Protocol for Amplification

1. Denature dsDNA with alkali:

Reagent	Quantity
Nuclease-free water	Up to 10 μL
10X Phi29 DNA Polymerase Reaction Buffer	1 μL
dsDNA	50 ng
NaOH (0.5M)	2 μL

2. Incubate at 30°C for 5 minutes. Place on ice for 1-2 minutes.
3. Add 1 μL of 1 M hydrochloric acid to neutralize.
4. Add 1-10 μL of 100μM-500μM primer. Incubate at 95°C for 3 minutes. Place on ice for 1-2 minutes.
5. Add the following components to the above reaction:

Reagent	Quantity
Nuclease-free water	Up to 50μL
Above reaction	12 – 21 μL
10X Phi29 DNA Polymerase Reaction Buffer	Up to 5 μL
dNTP(2mM)	5 – 7.5 μL
Phi29 DNA Polymerase(10U/μL)	1 μL

6. Incubate at 30°C for 2 hours.
7. Stop the reaction at 65°C for 10 minutes.

Notes

1. For research use only.