



## FIREPol® DNA Polymerase

Cat. No.	Pack Size	Conc.
01-01-0000S	100 U SAMPLE	5 U/μl
01-01-00500	500 U	5 U/μl
01-01-01000	1000 U	5 U/μl
01-01-02000	2000 U	5 U/μl

For *in vitro* use only

### Description:

FIREPol® is a highly processive, thermostable DNA polymerase. Due to its genetic modifications FIREPol® has an enhanced stability at room temperature with no activity loss for up to 1 month. The enzyme has 5'→3' polymerization-dependent exonuclease replacement activity but lacks 3'→5' exonuclease activity.

### Source:

Purified from an *E.coli* strain that carries an overproducing plasmid containing a modified gene of *Thermus aquaticus* DNA Polymerase.

### Applications:

- Suited for a wide range of PCR assays
- TA cloning

### Reagents Provided:

- **FIREPol® DNA Polymerase**
- **10x Reaction buffer B** (Mg<sup>2+</sup> free)  
*0.8 M Tris-HCl, 0.2 M (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 0.2% w/v Tween-20*
- **10x Reaction buffer BD** (Mg<sup>2+</sup> and detergent free)  
*0.8 M Tris-HCl, 0.2 M (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>*
- **25 mM MgCl<sub>2</sub>**
- **10x Solution S**  
*Additive that facilitates amplification of difficult templates (e.g. GC-rich DNA templates). This solution should be used at a defined working concentration (1x, 2x or 3x solution).*  
**Solution S is NOT a reaction buffer and should be used ONLY IF non-specific amplifications occur.**

### Concentration:

5 U/μl

### Unit definition:

One unit is defined as the amount of enzyme required to catalyze the incorporation of 10 nmol of dNTPs into an acid-insoluble form in 30 minutes at 74°C.

### Storage and Dilution buffer:

50% glycerol (v/v), 20 mM Tris-HCl pH 8.7 at 25°C, 100 mM KCl, 0.1 mM EDTA and stabilizers.

### Quality control:

The enzyme is free of nicking and priming activities, exonucleases and non-specific endonucleases. SDS/PAGE - 95 kD band, >98% pure. Activity and stability tested via thermo-cycling. The error rate per nucleotide per cycle is ~ 2.5 x 10<sup>-5</sup>; the accuracy is ~ 4 x 10<sup>4</sup>. Estimated half life at 95°C is 1.5 hours.

### Shipping and Storage conditions:

Routine storage: -20°C

Shipping and temporary storage for up to 1 month at room temperature has no detrimental effects on the quality of FIREPol® DNA Polymerase.

### Recommended PCR reaction mix:

Component	Volume	Final conc.
FIREPol® DNA Polymerase (5 U/μl)	0.4-1.0 μl	0.02-0.05 U/μl (2-5 U)
10x Buffer B or BD	10 μl	1x
25 mM MgCl <sub>2</sub>	6-10 μl	1.5-2.5 mM
20 mM dNTP mix	1 μl	200 μM
Primer Forward (10 pmol/μl)	1-3 μl	0.1-0.3 μM
Primer Reverse (10 pmol/μl)	1-3 μl	0.1-0.3 μM
DNA template	5-20 μl	5-100 ng/μl
10x Solution S <b>Not for standard PCR</b>	0, 10, 20 or 30 μl	1x, 2x or 3x
H <sub>2</sub> O PCR grade	Up to 100 μl	
<b>Total</b>	<b>100 μl</b>	

### Recommended PCR cycles:

Cycle step	Temp.	Time	Cycles
<b>Initial denaturation</b>	<b>95°C</b>	<b>3-5 min</b>	<b>1</b>
Denaturation	95°C	30-60 s	26-35
Annealing	50-68°C	30-60 s	
Elongation	72°C	1-4 min	
Final elongation	72°C	5-10 min	1

**IMPORTANT:** Annealing temperature should be 2-6°C lower than the primer melting temperature. Elongation time should be ~1 min/1 kb.

### Safety warnings and precautions:

This product and its components should be handled only by persons trained in laboratory techniques. It is advisable to wear suitable protective clothing, such as laboratory overalls, gloves and safety glasses. Care should be taken to avoid contact with skin or eyes. In case of contact with skin or eyes, wash immediately with water.

*Some applications this product is used in may require a license which is not provided by the purchase of this product. Users should obtain the license if required.*