DATA SHEET

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FastPol HF DNA Polymerase

High Fidelity DNA polymerase with enhanced processivity.
Pyrococus furiosus, recombinant, *E. coli*

Cat. Nº.	Amount
☐ POL-132XS	100 units
☐ POL-132S	250 units
■ POL-132M	500 units
☐ POL-132L	2 x 500 units
☐ POL-132XL	4 x 500 units

Unit Definition:
One unit is defined as the amount of the enzyme required to catalyze the incorporation of 10 nmoles of dNTPs into an acid-insoluble form in 30 minutes at 70 °C.

Concentration:
2 units/µL

Shipping:
Shipped on blue ice

Storage Conditions:
Store at -20 °C

For *in vitro* use only!

Additional Storage Conditions:
Avoid freeze/thaw cycles

Shelf Life:
24 months

Kit contents:

FastPol HF DNA Polymerase (blue cap)
2 units/µl FastPol DNA Polymerase in Tris-HCl, KCl, EDTA, DTT, 50% (v/v) Glycerol, pH 8.0 (25°C) and stabilizers.

FastPol HF Reaction Buffer complete (red cap) - 5x conc.
Optimized buffer for FastPol Polymerase.

Description:
FastPol HF is a designed highly thermostable DNA Polymerase that combines maximum fidelity and processivity in one. With an error rate 6x lower than that of Pyrococcus furiosus (Pfu) and extension rate up to 50x greater than Taq, FastPol HF DNA Polymerase generates improved product yields with high speed without compromising accuracy. The combined enhanced fidelity and processivity with high yields using minimum amount of enzyme make from FastPol an ideal choice for routine PCR, cloning and also long and difficult amplifications. FastPol HF is supplied with an optimized 5x buffer system containing Mg^{2+} and suitable for most applications.

* encoded by the same gene of Phusion®

Properties:

5' →3' Exonuclease: No
3' →5' Exonuclease: Yes
Product overhang: blunt
Recommended extension time: 15-30 sec/kb
GC-Rich samples: Yes
Long range PCR: up to 20 kbp

PCR Reaction Setup

The PCR procedure below shows appropriate volumes for a single 50-µL reaction. For multiple reactions, prepare a master mix of components common to all and then dispense appropriate volumes into each PCR reaction tube prior to adding template DNA and primers.

Thaw, mix, and briefly centrifuge each component before use.

Add the following components to a microcentrifuge tube:

1. Prepare PCR master mix
Note: Consider the volumes for all components listed next steps to determine the correct amount of water required to reach your final reaction volume.

Components	50 µL rxn	[final]
Water, grade PCR	To 50 µL	
5x Reaction Buffer	10 µL	1X
dNTP (Mix 10 mM)	1 µL	200 µM each
FastPol HF DNA Polymerase (2U/ µL)	0,5 µl	1 U/reaction

*Do not exceed 1U/50 µL reaction of FastPol

Mix and briefly centrifuge the components.

2. Add template DNA and primers

Components	50 µL rxn	[final]
Foward primer (10 µM)	1,5 - 5 µl	0,3 – 1 µM
Reverse primer (10 µM)	1,5 - 5 µl	0,3 – 1 µM
DNA template recommended		10 ng**

* The minimum recommended primer concentration is 0,3 µM . For maximum product yield a final concentration of 1 µM is used

**Genomic DNA: 10 ng - 1 µg; plasmidial or viral DNA: 5 pg - 10 ng

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3. Cycling instruction

Recommended cycling conditions:

Step		Temp.	Time
Initial denaturation		98 °C	2 min
25 - 35 cycles	Denaturation	98 °C	20 sec
	Annealing[1]	49-68 °C	15 sec
	Elongation	68 °C	15-30 sec/kb
Final extension		68 °C	1 - 10 min
Hold		4 - 8 °C	hold

[1] The annealing temperature depends on the melting temperature of the primers used.

For optimal specificity and amplification an individual optimization of the recommended parameters may be necessary for each new template DNA and/or primer pair.