

# PhilisaFAST® DNA Polymerase



**REF** 250024      **LOT** 80640995      2019-12-08

Size: 250 Units  
Concentration: 5 Units/ $\mu$ l  
Storage Temperature: -20 °C  
350639-7  
2017-12

**For Research Use Only. Not for use in diagnostic procedures.**

## DESCRIPTION

The PhilisaFAST DNA polymerase is a hot-start PCR enzyme specifically formulated for fast PCR. The speed of the enzyme facilitates extension times around 10 sec/kb, lowering the run time compared to conventional protocols. Hot-start of the enzyme is accomplished by disassociation of the monoclonal antibody during the initial DNA denaturation step, preventing mis-priming and primer-dimer formation before thermal cycling. Two types of reaction buffer are supplied. Fast Buffer I is recommended for use with rapid PCR cycling. Either Fast Buffer I or II may be used with standard ramp rates.

## COMPONENTS

PhilisaFAST DNA Polymerase (5 U/ $\mu$ L) 50 $\mu$ l  
10X Fast Buffer I (30mM MgCl<sub>2</sub>) 1ml  
10X Fast Buffer II (20mM MgCl<sub>2</sub>) 1ml  
dNTP Mixture (2.5mM each) 800 $\mu$ l  
Magnesium Chloride (25mM) 500 $\mu$ l

## Supplied dNTP Mixture (2.5mM each)

dNTP Mixture is ready for use in PCR without dilution.  
Form: Dissolved in water (sodium salts), pH 7-9  
Purity:  $\geq$  98% for each dNTP

One unit is the amount of enzyme that will incorporate 10nmol of dNTP into acid-insoluble products in 30 minutes at 74 °C with activated salmon sperm DNA as the template-primer.

## Reaction mixture for unit definition:

25mM TAPS (pH 9.3 at 25 °C)  
50mM KCL  
2mM MgCl<sub>2</sub>  
0.1mM DTT  
200 $\mu$ M each dATP•dGTP•dCTP  
100 $\mu$ M [<sup>3</sup>H] -dTTP  
0.25mg/mL activated salmon sperm DNA

## PURITY

Nicking, endonuclease and exonuclease activity were not detected after the incubation of 0.6 $\mu$ g of supercoiled pBR322, 0.6  $\mu$ g of  $\lambda$  DNA or 0.6 $\mu$ g of  $\lambda$ -Hind III digest with 10 units of this enzyme for 1 hour at 74 °C.

## APPLICATION

For DNA amplification by PCR, especially for rapid thermal cycling conditions.

## PCR PRODUCTS

As most PCR products amplified with PhilisaFAST DNA Polymerase will have one A added at 3'-terminus, the obtained PCR product can be directly used for cloning into T-vector. It is also possible to clone the product in blunt-end vectors after blunting and phosphorylation of the end.

**PCR Test:** Good performance of PCR amplification was confirmed by using  $\lambda$  DNA as a template (amplified fragment: 20kb).

## General reaction mixture for PCR (total 50 $\mu$ L):

10X Fast Buffer I or II 5 $\mu$ l  
dNTP Mixture (2.5 mM each) 4 $\mu$ l  
Template Variable, < 500ng  
Primer 1 0.2 - 1 $\mu$ M (final conc.)  
Primer 2 0.2 - 1 $\mu$ M (final conc.)  
PhilisaFAST DNA Polymerase (5 U/ $\mu$ L) 0.25 $\mu$ l  
Sterilized distilled water up to 50 $\mu$ l

## Note:

Optimal MgCl<sub>2</sub> concentration may be achieved by adjustment of the buffer amount (e.g., 0.8 - 1.2X) or supplementing with additional MgCl<sub>2</sub>.

## PCR CONDITIONS

(A) Using a rapid Streck platform  
95 °C 30 - 60 sec (Hot-Start)  
95 °C 5 - 10 sec  
55 °C - 65 °C 5 - 15 sec  
72 °C 1 - 2 sec / 100 bp } 30 - 45 cycles

(B) Using thermal cyclers with standard ramp rates  
95 °C 1 - 2 min (Hot-Start)  
95 °C 10 - 30 sec  
55 °C - 65 °C 10 - 30 sec  
72 °C 1 - 2 sec / 100 bp } 30 - 45 cycles

## Notes:

- For short amplicons, the 72 °C step may be omitted (2-step PCR)
- For annealing near 68 °C, use 2-step PCR with optimized hold time
- For GC-rich or long template/amplicon, the denaturation temp may be increased up to 98 °C

## ORDERING INFORMATION

Please call our Customer Service Department toll free 800-228-6090 for assistance. Additional information can be found online at [www.streck.com](http://www.streck.com).

## Note:

This product is for research use only. It is not intended for use in therapeutic or diagnostic procedures for humans or animals. This product may not be resold or transferred, modified for resale or transfer, or used to manufacture commercial products. For additional information please contact Streck at 800-843-0912 or online at [www.streck.com](http://www.streck.com).

## GLOSSARY OF SYMBOLS

See the Instructions (IFU) tab under Resources on the product page at [www.streck.com](http://www.streck.com).

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