Data sheet

FastPANGEA™-Long PCR DNA Polymerase

Catalog Number: P0060 (5U/µL)

Introduction

FastPANGEA™-Long PCR DNA Polymerase combines high quality recombinant Taq DNA Polymerase with a high-fidelity, proofreading polymerase. This enzyme blend has the 5'→3' exonuclease activity of Taq DNA polymerase as well as the 3'→5' exonuclease activity of the proofreading polymerase. Alone, Taq DNA polymerase is inefficient at amplifying fragments larger than 3–5kb due to its inability to repair nucleotide mismatches following misincorporation. The addition of a small quantity of proofreading enzyme allows mismatches to be repaired and extension to continue, resulting in the amplification of long amplicons with high yield. The presence of the proofreading polymerase significantly increases fidelity (6.5X) as compared to Taq polymerase alone. This mixture of enzymes allows for long and accurate PCR amplification of targets from a variety of templates, such as 5-15 kb of genomic DNA. The enzyme blend generates PCR products whose ends are compatible with either blunt-end or TA cloning procedures with A-tailed ends favored over blunt ends in an approximately 3:1 ratio.

Features

- Extreme Fidelity
- Robust Reactions
- High Speed
- High Yield
- Versatile

KIT CONTENTS

Item	Quantity
FastPANGEA™-Long PCR DNA Polymerase	**200U**
Buffer PA (10x)	**1.5 mL**
DMSO (100%)	**50 µL**
$MgCl_2$ (25 mM)	**1.5 mL**

Storage: Store at -20 °C.

Quality Certifications

- Functionally tested in PCR.
- Undetected bacterial DNA (by PCR).

Applications:

- PCR-Cloning: highly recommended for cloning into pSpark® DNA cloning vectors.
- Primers extension.
- Long or difficult amplification.
- High-Throughput PCR.

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BASIC REACTION CONDITIONS FOR PCR AMPLIFICATIONS

Carefully centrifuge all tubes before opening to improve recovery. PCR reactions should be set up on ice. Prepare a mix for the appropriate number of samples to be amplified.

The following protocol is recommended for a 20 µl reaction volume:

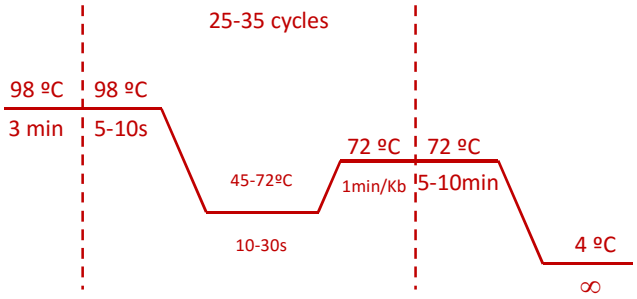
1. Assemble the following reagents in a thin-walled PCR tube.

Component	Volume reaction 20 µL	Final concentration
Primer A	X µL	0.75 µM[(1)]
Primer B	X µL	0.75 µM[(1)]
Template DNA	X µL	20-50 ng DNA[(2)]
DMSO (optional)	0.6 µL	3%[(3)]
8 mM dNTPs	2 µL	0.8 mM
Buffer PA (10x)	2 µL	1X
FastPANGEA™-Long PCR DNA Polymerase	0.2 µL	0.05U/µL
$MgCl_2$ (25 mM)	2µL	2.5mM
Nuclease-Free Water to a final volume of	20 µL	

[(1)]The recommendation for final primer concentration is 0.5 µM but it can be varied in a range of 0.2-1.0 µM if needed.
[(2)]For gDNA used 100-300 ng DNA.
[(3)]Addition of DMSO is recommended for GC-rich amplicons. **If DMSO is added in the PCR reaction, Tm must be decreased about 3º C.**

2. Mix reagents completely, and then transfer to a thermocycler.

3. Perform the following cycling conditions:



- ✓ *As with all PCR reactions, conditions may need to be optimized to achieve maximum amplicon yield. You may be able to adjust your PCR conditions to optimize reaction.*
- ✓ *Genomic targets over 20kb may require additional optimization.*

PRODUCT USE LIMITATION

This product is developed, designed and sold exclusively for research purposes and in vitro use only. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals. Please refer to www.canvaxbiotech.com for Material Safety Data Sheet of the product.

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2X PANGEA-Long PCR Master Mix

Catalog Number: P0022 (200 rxn)

Introduction

PANGEA-Long PCR Master Mix combines Taq DNA Polymerase and a DNA proofreading polymerase with 3' to 5' exonuclease activity that is optimized for PCR amplification of very long DNA templates (long range PCR). Alone, Taq DNA polymerase is inefficient at amplifying fragments larger than 3–5kb due to its inability to repair nucleotide mismatches following misincorporation. The addition of a small quantity of proofreading enzyme allows mismatches to be repaired and extension to continue, resulting in the amplification of long amplicons with high yield. The presence of the proofreading polymerase significantly increases fidelity (6.5X) as compared to Taq polymerase alone. This mixture of enzymes allows for long and accurate PCR amplification of targets from a variety of templates, such as 5-15 kb of genomic DNA. PANGEA Long polymerase generates long templates with an accuracy and speed previously unattainable with other thermostable DNA polymerases. PANGEA-Long DNA Polymerase Master Mix possesses 3´→ 5´ exonuclease activity and it generates PCR products with blunt ends and generate 3´-adenine overhang in amplified DNA and thus such Taq amplified DNA could be cloned into T-vectors.

Features

- Extreme Fidelity
- Robust Reactions
- High Speed
- High Yield
- Versatile

KIT CONTENTS

Item	Quantity
2X PANGEA-Long PCR master mix *Includes: Mix DNA Polymerases, dNTPs, $MgCl_2$*	**2 ml**
DMSO	**50 µL**
$MgCl_2$ (25 mM)	**100 µL**

Separate tubes of DMSO and 25 mM $MgCl_2$ solutions are provided for further optimisation.

Storage: Store at -20 °C.

Quality Certifications

- Functionally tested in PCR.
- Undetected bacterial DNA (by PCR).

Applications:

- PCR-Cloning: highly recommended for cloning into pSpark® DNA cloning vectors.
- Primers extension.
- Long or difficult amplification.
- High-Throughput PCR.

(Continued on reverse side)

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The following protocol is recommended for a 20 µl reaction volume:

1. Assemble the following reagents in a thin-walled PCR tube.

Component	Volume reaction 20 µL	Final concentration
Primer A	X µL	0.75 µM[(1)]
Primer B	X µL	0.75 µM[(1)]
Template DNA	X µL	20-50 ng DNA[(2)]
DMSO (optional)	(X µL)	3%[(3)]
2X Polymerase master mix	10 µL	1X
Nuclease-Free Water to a final volume of	20 µL	

[(1)]The recommendation for final primer concentration is 0.5 µM but it can be varied in a range of 0.2-1.0 µM if needed.
[(2)]For gDNA used 100-300 ng DNA.
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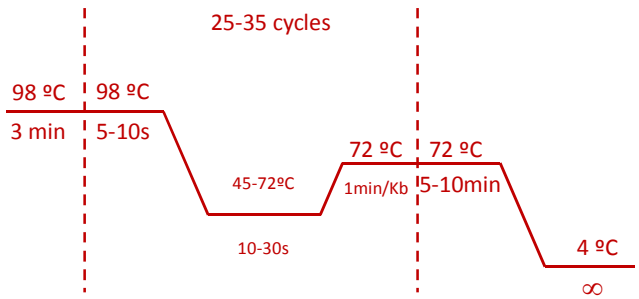

- ✓ *As with all PCR reactions, conditions may need to be optimized to achieve maximum amplicon yield. You may be able to adjust your PCR conditions to optimize reaction.*
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