Data sheet

PANGEA- High Fidelity DNA polymerase

Catalog Number: P0031 (50 units)
Catalog Number: P0032 (100 units)
Catalog Number: P0033 (500 units)

Introduction

PANGEA- High Fidelity DNA polymerase is a second generation high-fidelity DNA polymerase that offers extreme performance for all PCR applications. PANGEA DNA polymerase generates long templates with an accuracy and speed previously unattainable with other thermostable DNA polymerases. The error rate of **PANGEA DNA polymerase** is at least 50 fold lower than that of Taq DNA polymerase.

PANGEA DNA polymerase possesses the 5'→3' DNA polymerase activity, 3´→ 5´ exonuclease activity and it generates PCR products with blunt ends. PANGEA High Fidelity DNA polymerase is also suitable for amplification of long amplicons such as 10-15 kb of genomic DNA.

Features

- ✓ **Extreme Fidelity**
- ✓ **Robust Reactions**: maximal success with minimal optimization.
- ✓ **High Speed**: **PANGEA- High Fidelity DNA polymerase** extension times are 15-30 seconds/kb.
- ✓ **High Yield**: increased product yield using minimal amount of enzyme.
- ✓ **Versatile**: Can be used for routine PCR as well as long or difficult templates.

KIT CONTENTS

Item	P0031	P0032	P0033
PANGEA- High Fidelity DNA polymerase (2U/µl)	50 U	100 U	500 U
2.5X Buffer Uni	250 µL	500 µL	2x1.5 mL
DMSO*	50 µL	100 µL	500 µL
$MgCl_2$ (25 mM)*	50µL	100 µL	500 µL

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

Storage: Store at -20 °C.

Unit definition: One unit is defined as the amount of enzyme that incorporates 10 nmoles of dNTP's into acid-insoluble form in 30 minutes at 75°C under **assay conditions:** *25 mM TAPS-HCl, pH 9.0 (at 25°C), 100 mM KCl, 1.5 mM MgCl2 , 1 mM Beta-mercaptoethanol, 200 µM each dNTP, and 10 µg activated calf thymus DNA in 50 µl.*

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)

Distributed by:
Lab Unlimited
CARL STUART GROUP

Tallaght Business Park
Whitestown, Dublin 24,
Ireland
D24 RFK3

Tel: (01) 4523432
Fax: (01) 4523967
E-mail: info@labunlimited.com
Web: www.labunlimited.com

Quatro House, Frimley Road,
Camberley,
United Kingdom
GU16 7ER

Tel: 08452 30 40 30
Fax: 08452 30 50 30
E-mail: info@labunlimited.co.uk
Web: www.labunlimited.co.uk

canvax

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 50 µl reaction volume:

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

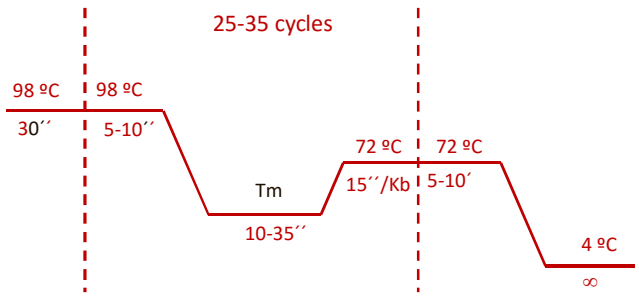
Component	Volume reaction 50 µL	Final concentration
Primer A	X µL	0.75 µM[1]
Primer B	X µL	0.75 µM[1]
Template DNA	X µL	20-50 ng DNA[2]
DMSO (optional)	1.5 µL	3%[3]
10 mM dNTPs	1 µl	0.2 mM each [4]
2.5x Buffer Uni	20 µl	1X [5]
PANGEA- High Fidelity DNA polymerase	0.5 µL	0.02 U/µL
Nuclease-Free Water to a final volume of	Up to 50 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 100-250 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.**
[4] For most of applications 200mM of each of dNTP's as final concentration is an optimal. It's not necessary to optimize dNTP's concentration. dUTP or other dUTP derivatives should be used replacing TTP in PCR reaction for "anti-contamination" PCR.
[5] 2.5X Uni Buffers provides very high reproducibility across the wide range of amplification conditions, including "fast-PCR" (reduced time of PCR reaction). 2,5X Uni Buffer contains 1.5mM Mg2+, as the final concentration. In some cases we recommend to optimize Mg concentration in the range 1.5-4.5mM

Note! It is critical that the PANGEA- High Fidelity DNA polymerase is the last component added to the PCR mixture, since the enzyme exhibits 3'->5' exonuclease activity that can degrade primers in the absence of dNTPs.

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.*
- ✓ *Extension time depends on amplicon length and complexity:*
  - *For low complexity DNA (eg. Plasmid, lambda, or BAC DNA) use 15 s per Kb.*
  - *For high complexity DNA (eg. gDNA) use 30s per kb. Do not exceed 1 min. per kb for amplicons that are <3 kb.*

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.

Distributed by:
Lab Unlimited
CARL STUART GROUP

Tallaght Business Park
Whitestown, Dublin 24,
Ireland
D24 RFK3
Tel: (01) 4523432
Fax: (01) 4523967
E-mail: info@labunlimited.com
Web: www.labunlimited.com

Quatro House, Frimley Road,
Camberley,
United Kingdom
GU16 7ER
Tel: 08452 30 40 30
Fax: 08452 30 50 30
E-mail: info@labunlimited.co.uk
Web: www.labunlimited.co.uk

Data sheet

2x PANGEA- High Fidelity DNA polymerase (Master Mix)

Catalog Number: P0061

500 x 20 µl reactions
200 x 50 µl reactions

Introduction

PANGEA- High Fidelity DNA polymerase is a second generation high-fidelity DNA polymerase that offers extreme performance for all PCR applications.

Ready-to-use 2X master mix preserves the fidelity and the yield in the reaction when using extremely short PCR protocols. Additionally, the user only needs to add template and primers minimizing the number of pipetting steps.

PANGEA DNA polymerase possesses the 5'→3' DNA polymerase activity, 3´→ 5´ exonuclease activity and it generates PCR products with blunt ends.

Features

- ✓ **Extreme Fidelity**
- ✓ **Robust Reactions**: maximal success with minimal optimization.
- ✓ **High Speed**: **PANGEA- High Fidelity DNA polymerase** extension times are 15-30 seconds/kb.
- ✓ **High Yield**: increased product yield using minimal amount of enzyme.

KIT CONTENTS

Item	P0061
2X PANGEA- High Fidelity DNA polymerase (Master mix)*	4 x 1.25 mL
DMSO**	500 µL
MgCl2 (25 mM)**	100 µL

**2X PANGEA- High Fidelity DNA polymerase Master Mix contains: PANGEA- High Fidelity DNA polymerase, 2X reaction buffer, dNTPs, and MgCl2.*
***Separate tubes of DMSO and 25 mM MgCl2 solutions are provided for further optimisation.*

Storage: Store at -20 °C.

Unit definition: One unit is defined as the amount of enzyme that incorporates 10 nmoles of dNTP's into acid-insoluble form in 30 minutes at 75°C under **assay conditions:** *25 mM TAPS-HCl, pH 9.0 (at 25°C), 100 mM KCl, 1.5 mM MgCl2 , 1 mM Beta-mercaptoethanol, 200 µM each dNTP, and 10 µg activated calf thymus DNA in 50 µl.*

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.
- ✓ High-Throughput PCR.

(Continued on reverse side)

Distributed by:
Lab Unlimited
CARL STUART GROUP

Tallaght Business Park
Whitestown, Dublin 24,
Ireland
D24 RFK3

Tel: (01) 4523432
Fax: (01) 4523967
E-mail: info@labunlimited.com
Web: www.labunlimited.com

Quatro House, Frimley Road,
Camberley,
United Kingdom
GU16 7ER

Tel: 08452 30 40 30
Fax: 08452 30 50 30
E-mail: info@labunlimited.co.uk
Web: www.labunlimited.co.uk

canvax

BASIC REACTION CONDITIONS FOR PCR AMPLIFICATIONS

Carefully centrifuge the tubes before opening to improve recovery.
PCR reactions should be set up on ice.

The following protocol is recommended:

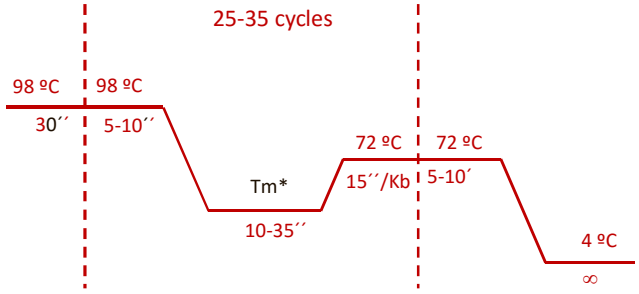
1. Assemble the following reagents in a thin-walled PCR tube.

Component	Volume reaction		Final concentration
	20 µL	50 µL	
Primer A	X µL	X µL	0.75 µM[(1)]
Primer B	X µL	X µL	0.75 µM[(1)]
Template DNA	X µL	X µL	20-50 ng DNA[(2)]
DMSO (optional)	0.6 µL	1.5 µL	3%[(3)]
PANGEA- High Fidelity DNA polymerase (2X)	10 µL	25 µL	1X
Nuclease-Free Water to a final volume of	Up to 20 µL	Up to 50 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 100-250 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.*
- ✓ *Extension time depends on amplicon length and complexity:*
  - *For low complexity DNA (Plasmid, lambda, or BAC DNA) use 15 s per Kb.*
  - *For high complexity DNA (gDNA) use 30s per kb. Do not exceed 1 min. per kb for amplicons that are <3 kb.*

*As a basic rule, for primers > 20 nt, anneal for 10–30 seconds at a Tm +3 °C of the lower Tm primer. For primers ≤ 20 nt, use an annealing temperature equal to the Tm of the lower Tm primer. If necessary, use a temperature gradient to find the optimal annealing temperature for each template-primer pair combination.

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.

Distributed by:
Lab Unlimited
CARL STUART GROUP

Tallaght Business Park
Whitestown, Dublin 24,
Ireland
D24 RFK3
Tel: (01) 4523432
Fax: (01) 4523967
E-mail: info@labunlimited.com
Web: www.labunlimited.com

Quatro House, Frimley Road,
Camberley,
United Kingdom
GU16 7ER
Tel: 08452 30 40 30
Fax: 08452 30 50 30
E-mail: info@labunlimited.co.uk
Web: www.labunlimited.co.uk