A&A BIOTECHNOLOGY
innovating life science

Manual

InfiniTaq™ PCR Kit

Long-range PCR Kit for amplification of DNA fragments up to 30 kb with high fidelity.

catalog #	size
1200-100	100 reactions in 25 µl
1200-1000	1000 reactions in 25 µl

For research use only.

Guarantee
A&A Biotechnology provides a guarantee on this product.
The company does not guarantee the correct performance of this kit in the event of:

- not adhering to the supplied protocol
- use of not recommended equipment or materials
- use of other reagents than recommended or which are not a component of the product
- use of expired or improperly stored product or its components

Table of Contents

Specification

amplification range	• human genomic DNA: up to 20 kb • Lambda DNA: up to 30 kb
proofreading	3'→5' exonuclease activity (proofreading activity)
resulting ends	blunt ends
extension time	60 s / 1 kb
applications	• NGS library construction • PCR, RT-PCR • cloning

Advantages

- Amplification of long DNA fragments up to 30 kb.
- 4x higher fidelity than *Taq* DNA polymerase.

Description

InfiniTaq™ PCR Kit contains a unique polymerase with an addition of PCR anti-inhibitors and enhancers and also reaction mix with nucleotides. Supplied **InfiniTaq™ PCR Mix** provides an ideal environment for **InfiniTaq™** polymerase to obtain the highest yields of long PCR products up to 30 kb.

Contents

component	1200-100	1200-1000	storage
InfiniTaq™ polymerase	100 µl	10 x 100 µl	-20 °C
2x InfiniTaq™ PCR Mix	2 x 1 ml	20 x 1 ml	-20 °C
50 mM $MgSO_4$	1 ml	2 x 1 ml	-20 °C
ultrapure water	1.5 ml	10 x 1.5 ml	-20 °C

InfiniTaq™ PCR Mix composition

component	final concentration
$MgSO_4$	2.5 mM
dNTPs	25 mM each dNTPs
buffer and stabilizers	

Additional equipment and reagents

- 0.2 ml PCR tubes
- thermocycler
- vorteks
- microcentrifuge

Important notes

- All solutions should be thawed thoroughly on ice, gently mixed by inverting the tube and briefly centrifuged before use.
- Activity of this product is not affected by up to 7x freeze-thaw cycles.
- We recommend testing for the optimal Mg^{2+} concentration. Follow chapter Troubleshooting.

Example PCR protocol

1. Thaw all components on ice. Gently vortex and briefly centrifuge. Place the tubes again on ice.

2. Place PCR tubes on ice or cool block and add:

component	25 µl reaction volume	final concentration
2x InfiniTaq™ PCR Mix	12.5 µl	–
50 mM $MgSO_4$	0 µl /0.25 µl Mg2+optimization	2.5 mM/3.0 mM
10 µM forward primer	0.5-1 µl	0.2-0.4 µM
10 µM reverse primer	0.5-1 µl	0.2-0.4 µM
InfiniTaq™ polymerase	0.3 µl	2 U
template DNA	variable	50-1000 ng
ultrapure water	up to 25 µl	–

3. Gently mix to avoid creating bubbles. Do not vortex. Centrifuge briefly if needed.

4. Place the tubes in the thermocycler and start the PCR program:

step	temperature	time	cycles
initial denaturation	95 °C	5 min	1
denaturation	95 °C	15 s	30
annealing	60-72 °C*	30 s	
extension	65 °C	30-60 s / 1 kb**	
final extension	72 °C	10 min	1

*Use primers optimized for the specific annealing temperature range.
We recommend setting the extension time to **60 s/1 kb for fragments longer than **5 kb**.

5. Store the PCR product at temp. from -20 °C up to 4 °C.

Troubleshooting

Mg^{2+}optimization

The provided **2x InfiniTaq™ PCR Mix** is equipped with Mg^{2+} ions at concentrations suitable for most amplicons. Nevertheless, we advise assessing the optimal Mg^{2+} concentration for each new amplicon, as this may lead to significant improvements in results.

- **2.5 mM** - reaction performed using the basic blend **2x InfiniTaq™ PCR Mix / 25 µl** reaction volume.
- **3.0 mM -** reaction performed using the basic blend **2x InfiniTaq™ PCR Mix** with **0.25 µl 50 mM $MgSO_4$ / 25 µl** reaction volume.

No PCR product

If the PCR product could not be obtained, please try the following:

- Lower the annealing temperature by 5 °C.
- Keep increasing the template DNA amount by 100 ng, up to maximum 1000 ng.
- Check the quality of template DNA by gel electrophoresis.

Non-specific PCR products

If a smear or non-specific PCR products are observed during electrophoresis, please try the following:

- Increase the annealing temperature by 5 °C.
- Decrease primers concentration by half.
- Decrease the amount of cycles by 5.
- Keep decreasing the template DNA amount by 100 ng, down to minimum 100 ng.

A&A BIOTECHNOLOGY

innovating life science

A&A Biotechnology, ul. Strzelca 40, 80-299 Gdańsk, Poland
phone +48 883 323 761, +48 600 776 268
info@aabiot.com, www.aabiot.com

version 2024-1