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Manual

RUN 5 DNA polymerase

Taq DNA polymerase with the reaction buffer. Concentration 5 U/µl.

catalog #	size
1001-200-5	200 U
1001-1000-5	1000 U

For research use only.

Guarantee

A&A Biotechnology provides guarantee on this product.
The company does not guarantee 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

Advantages

- *Taq* DNA polymerase is the most popular DNA polymerase in PCR procedures.
- The enzyme is recommended for routine PCR reactions.

Description

RUN 5 DNA polymerase is *Taq* polymerase purified from *E. coli* strain carrying a plasmid with a cloned gene encoding a DNA polymerase from *Thermus aquaticus*. Enzyme catalysis incorporation of deoxynucleotides to 3' end of dsDNA at temperature 70-80 °C and presence of Mg^{2+} ions.
Taq DNA polymerase lacks 3'-5' exonuclease activity, but possesses weak 5'-3' exonuclease activity.

Contents

	1001-200-5	1001-1000-5	storage
RUN 5 polymerase storage buffer: 100 mM KCl, 50 mM Tris-HCl pH 8.5, 0.5% Tween, 0.5% Triton X-100, 50% glicerol (v/v).	200 U	1000 U	-20 °C
RUN reaction buffer 10x PCR reaction buffer: 100 mM KCl, 100 mM $(NH_4)_2SO_4$, 200 mM Tris-HCl pH 8.5, 20 mM $MgSO_4$, 1% Igepal.	1 x 1.5 ml	4 x 1.5 ml	-20 °C

Unit definition

One unit of the enzyme catalyzes the incorporation of 15 nmol of dNTP into a polynucleotide fraction in 30 min at 75 °C.

Notes

- Before using, thoroughly thaw and gently mix by inverting the tubes.

PCR protocol

1. Thaw all components on ice, gently mix by inverting the tubes and briefly centrifuge. Place the tubes on ice again.

2. Place PCR tubes on ice and add:

component	volume 50 µl	final concentration
RUN reaction buffer	5 µl	1X
dNTP Mix (10 mM)	1-1.25 µl	200-250 µM
Primer 1 (10 µM)*	1 µl	0.2 µM
Primer 2 (10 µM)*	1 µl	0.2 µM
RUN 5 polymerase	0.25 µl	1.25 U
DNA template	variable	10 pg -1 µg
ultrapure water	up to 50 µl	

*For optimization, a primer titration should be performed from 0,2 µM do 1 µM final concentration.

3. Gently mix the samples and briefly centrifuge.

4. Place the tubes in the thermocycler and start the PCR programme.
An example amplification profile:

reaction step	temperature	time	number of cycles
initial denaturation	95 °C	2-3 min	1
denaturation	95 °C	15 s	40
annealing*	50-68 °C	30 s	
extension**	72 °C	30 s	

*Annealing temperature depends on primer sequence and the composition of the reaction mixture.
**Time of extension depends on the length of the amplicon.

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