Heat-Activated DNA Polymerase

Catalogue No.:abx461024

Abbexa's Heat-Activated DNA Polymerase is a heat-activated thermostable enzyme with improved specificity as compared to standard polymerases, by eliminating all non-specific priming and the formation of primer-dimers. It is inactive at room temperature, allowing for reaction set-up at room temperature, and therefore requires activation by heat treatment for 10 minutes prior to PCR cycling. The enzyme is available as ready-to-use 2X reaction mixes: abx461025, abx461026.

Contents:

Component	250 U	500 U	5 kU
Heat-Activated DNA Polymerase	50 µl	100 µl	10 × 100 µl
10X Reaction Buffer	1.2 ml	2 × 1.2 ml	20 × 1.2 ml
50 mM $MgCl_2$ Solution	1.2 ml	1.2 ml	10 × 1.2 ml

This product does not contain dNTPs - this is available for purchase separately: abx461012

Target: Heat-Activated DNA Polymerase

Tested Applications: PCR

Form: Liquid

Storage: Store all components at -20 °C. It is not recommended to store the enzyme at -80 °C as ice crystals may form on the active site, which can affect the enzyme activity. Avoid repeated freeze/thaw cycles.

Validity: Up to 12 months.

Buffer: The exact formulation is proprietary.

Biological Activity: One unit is defined as the amount of enzyme that incorporates 10 nmol of dNTPs into acid-insoluble form in 30 minutes at 72 °C.

Concentration: 5 U/µl

Note: THIS PRODUCT IS FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC, THERAPEUTIC OR COSMETIC PROCEDURES. NOT FOR HUMAN OR ANIMAL CONSUMPTION.

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Directions for use:

Reaction Components:

Component	Volume
Template and Primers	Variable, as required
50 mM $MgCl_2$ Solution	3 µl
10X NH_4 Reaction Buffer	5 µl
100 mM dNTP Mix (abx461012)	0.5 µl
Heat-Activated DNA Polymerase	1 µl
Water	Variable, up to 50 µl
Total Volume	**50 µl**

Thermal Cycling Conditions:

- Activation: pre-heat at 95 °C for 10 minutes
- Denaturation: 94-96 °C
- Annealing: dependent on primer T_m
- Extension: 72 °C, allowing 15-30 seconds per kb

Notes:

- Pre-incubate at 95 °C for 10 minutes to activate the enzyme. Subsequently, the reaction can be treated according to the end user's existing protocol.
- If extension time exceeds 2.5 minutes, a maximum of 30 cycles should be used. Increasing the number of cycles may lead to smearing when run on an agarose gel.
- The optimal conditions will vary from reaction to reaction and are dependent on the system used. Each parameter needs to be adjusted by the end user and some optimization may be required.
- The optimal final Mg^{2+} concentration is likely to be in the region of 1.5 - 2.5 mM.
- It is recommended to use at least 1 U of Heat-Activated DNA Polymerase per 50 µl reaction volume.

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