Check the product label for actual catalog number, lot and expiry date.

ALLin™ Bst DNA Polymerase

CAT.#	SIZE	COMPONENTS	COMPONENT COMPOSITION
IDE0201	8000 u	8000 u – ALLin™ Bst DNA Polymerase, 8 u/µl 2 x 1.25 ml – 10X ALLin™ Bst Buffer 3 x 1.7 ml – 5X Bst Enhancer	Glycerol-free formulation of the recombinant Bst DNA Polymerase (large fragment) in storage buffer. Excellent for both common and lyophilization-ready workflows. ALLin™ Bst Buffer contains optimal concentration of dNTPs and magnesium sulphate as well as stabilizers. The proprietary Bst Enhancer accelerates the reaction.

Storage *In the dark at -20°C. As the enzyme is glycerol-free, to avoid multiple freezing-thawing, aliquot the enzyme into 5-10 sterile tubes.*

APPLICATIONS

- Isothermal DNA amplification at elevated temperature
- Real-time detection of DNA amplification
- MDA – multiple displacement amplification
- LAMP - loop-mediated isothermal amplification
- WGA - whole genome amplification
- RAM - ramification & RPA - recombinase polymerase ampl.

BENEFITS

- Glycerol-free formulation of robust Bst DNA Polymerase, supplied with all-included buffer with dNTPs and proprietary reaction enhancer
- Active at high temperatures in a range of 55-70°C
- Fast DNA amplification at constant temperature
- Ideal for complex templates and crude samples
- Ensures <5 molecules LOD (limit of detection)

PRODUCT DETAILS

The ALLin™ Bst DNA Polymerase is a recombinant protein, representing a large fragment of the *B. stearothermophilus* DNA Polymerase expressed in *E. coli* cells. This robust polymerase with a strong strand displacement activity and high temperature tolerance ensures high amplification yield at constant temperature when working with impure or low-copy number targets as well as with complex templates. The Allin™ Bst Buffer includes optimal concentrations of magnesium and dNTPs, what minimizes pipetting steps. This ALLin™ Bst DNA Polymerase-buffer system together with a supplied enhancer, detects <5 DNA targets in a short time without the use of a thermocycler.

This glycerol-free Bst DNA Polymerase is also an excellent tool for development of dry format lyophilized kits for pathogen detection using isothermal amplification techniques.

For more convenience, we recommend our kit formulations: ALLin™ Isothermal DNA Amplification Kit and ALLin™ Isothermal 1Step RNA Amplification Kit, both including a master mix with optimized high-performance buffer.

PERFORMANCE

Technical characteristics of Bst DNA Polymerase (large fragment):

- Strong 5′ - 3′ strand displacement activity
- Strong 5′ - 3′ polymerase activity
- No 5′ - 3′ exonuclease activity
- No 3′ - 5′ exonuclease (proofreading) activity
- Only minor reverse transcriptase activity
- Optimal amplification temperature is 65°C
- Working temperature range is 55 - 70°C
- Optimal reaction time is 20 minutes (depends on the buffer)
- If needed, the reaction can be extended to 30 - 60 minutes
- The enzyme is inactivated in 10 minutes at 80°C

The use of this product in certain applications may be covered by patents of third parties. The user has to analyse all applicable Limited Use Label Licenses and may need licensing from third parties.

ISOTHERMAL AMPLIFICATION PROTOCOL EXAMPLE

- Take typical measures to prevent contamination, keep your bench clean, wear gloves, use sterile tubes and filter pipet tips.
- Include a no-template control and positive controls in parallel.
- Thaw and keep reagents on ice. Mix well before use.
- Perform the reaction at 65°C. If needed, optimize the reaction temperature between 55 - 70°C for each template/primers system. Complex templates may require higher temperature.
- Suggested reaction time is 20 - 30 minutes. For some low copy number targets 30 - 60 minutes might be required.
- Design primers with predicted melting temperature of about 60°C.
- Prepare 10X primer mix in water or TE Buffer, for example, for LAMP: 16 µM FIP, 16 µM BIP, 2 µM F3, 2 µM B3, 8 µM LoopF, 8 µM LoopB.

✓ Prepare a 25 µl reaction:

10X ALLin™ Bst Buffer	2.5 µl
5X Bst Enhancer	5 µl
10X primer mix	2.5 µl
Template DNA	1 µl
PCR Water	to 24 µl
ALLin™Bst DNA Polymerase, 8 u/µl	1 µl

✓ Mix gently, avoid bubbles.

✓ Place into the thermostat or qPCR instrument to incubate:

Amplification	65°C – 20 - 30 min
temperature can be between 55-70°C, time between 20 - 60 min	
Optional: Inactivation	80°C - 10 min

Store reactions for short time on ice, for long time at -20°C.
High yield amplification product may increase the viscosity of the solution, mix well and dilute if needed for downstream applications.

IN VITRO RESEARCH USE ONLY

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