ArcticZymes Technologies

IsoPol® Isothermal polymerases

For point-of-care diagnostics assays

www.arcticzymes.com

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In point-of-care (POC) settings, speed is essential to provide actionable information for rapid implementation of disease management, which is why isothermal methods demand high quality polymerases with unique features. ArcticZymes introduces unique isothermal polymerases (IsoPol®) carefully engineered to improve decisive features as strand displacement, processivity and tolerance to biological materials.

Efficient strand displacement allows the polymerase to amplify fast while displacing the second DNA strand during synthesis. High processivity ensures that the polymerase can amplify more nucleotides consecutively without dissociation from the DNA template. And finally, higher tolerance to salt and biological matrices introduces opportunities for simplifying or omitting sample prep steps before starting the test.

Taken together, the engineered isothermal polymerases from ArcticZymes allows faster diagnostics in challenging matrices using popular diagnostic methods such as loop-mediated amplification (LAMP).

IsoPol® BST⁺

IsoPol® SD⁺

IsoPol®

- IsoPol BST⁺ is an in silico designed homologue of Bst DNA Polymerase (large fragment) suitable for amplification at elevated temperatures with an optimum at 65°C.
- IsoPol BST⁺ and IsoPol SD⁺ are engineered for enhanced amplification performance and higher inhibitor tolerance.
- IsoPol SD⁺ and IsoPol DNA Polymerase are ideal for amplification at temperatures above 20°C and 37°C.

IsoPol® – For point-of-care diagnostics assays

Strand displacement and processivity

Isothermal amplification techniques are under rapid development as an alternative to conventional PCR in amplification of nucleic acids, especially for detection of pathogens. Conventional PCR methods uses repetitive cycling temperatures when amplifying, while isothermal amplification techniques occur at a single and fixed temperature while still allowing fast, and exponential amplification. Maintaining a single temperature allows the use of simpler, more portable, and robust instruments compared to conventional thermocyclers, making isothermal methods well suited for use in point-of-care diagnostics.

Although there are several different isothermal amplification techniques, two key requirements for effective assay performance are using a DNA polymerase with excellent strand displacement activity and high processivity.

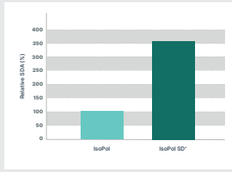


Fig. 1 IsoPol SD⁺ shows superior strand displacement activity compared to IsoPol polymerase.

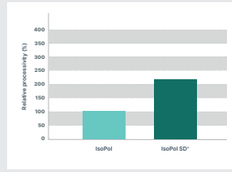


Fig. 2 IsoPol SD⁺ is engineered for improved processivity. Processivity was determined at 37°C using a primed circular ssDNA.

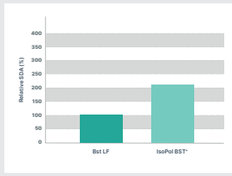


Fig. 3 IsoPol BST⁺ has superior strand displacement activity compared to Bst LF. SDA was measured at 37°C using a nicked dsDNA template.

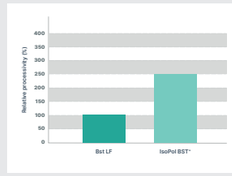


Fig. 4 IsoPol BST⁺ has enhanced processivity compared to Bst LF. Processivity was determined at 37°C using a primed circular ssDNA.

Improved strand displacement and processivity by engineering. The proprietary engineering introduced to the IsoPol and Bst large fragment (LF) polymerase backbones to make IsoPol BST⁺ and IsoPol SD⁺, respectively, improved both the strand displacement and processivity more than two-fold. IsoPol SD⁺ is ideal for amplifications occurring at temperatures up to 42°C.

Isothermal Amplification

The improved strand displacement and processivity of ArcticZymes IsoPol BST⁺ polymerase significantly improved amplification speed and efficiency in LAMP and RT-LAMP.

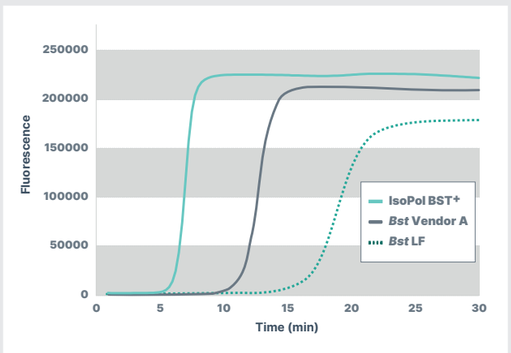


Fig. 5 IsoPol BST⁺ provides faster amplification in LAMP

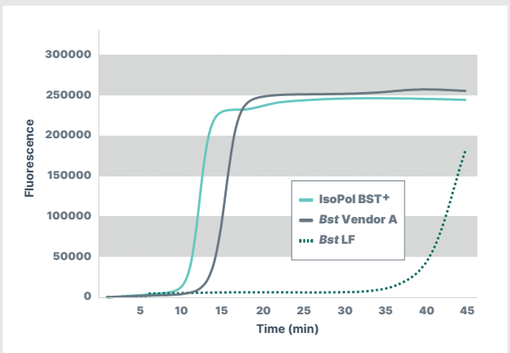


Fig. 6 provides superior time-to-results in RT-LAMP

IsoPol BST⁺ results in faster detection of target in both LAMP and RT-LAMP.
When IsoPol BST⁺ was used in LAMP and RT-LAMP, time to results were strongly reduced compared to the non-engineered backbone, Bst LF. IsoPol BST⁺ also significantly outperformed engineered versions of Bst LF from another vendor (Bst Vendor A). LAMP was used to detect 0.05 ng of λ DNA (Fig 5). RT-LAMP was performed with AMV RT to detect 10 ng of MS2 RNA template (Fig 6). LAMP and RT-LAMP were performed at 65°C.

In addition to strand displacement activity and processivity, inhibitor tolerance and sample size are of particular importance in point-of-care diagnostics.

Inhibitor tolerance

Minimally processed samples that contain impurities such as serum or salt can interfere or inhibit the polymerase amplification process.

IsoPol BST⁺ and IsoPol SD⁺ were specifically designed to tolerate high salt concentrations.

LAMP was performed using different serum concentrations in the sample matrix (Fig 7). IsoPol BST⁺ provided robust amplifications across several serum concentrations demonstrating high tolerance to matrix effects in LAMP compared to an engineered Bst version from another vendor.

Broad salt tolerance also allows for flexibility where multiple enzymes with distinct buffer conditions are required, such as with HDA or NEMA.

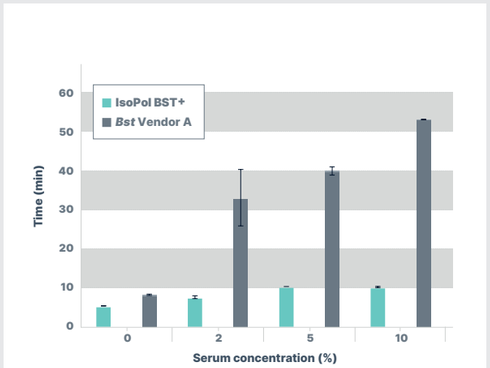
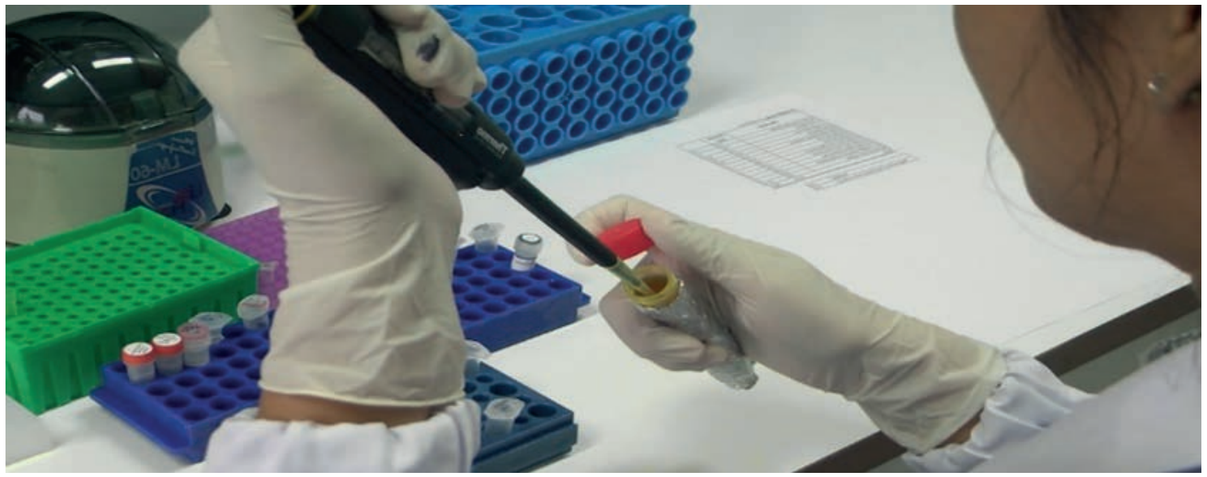


Fig 7. PCR master mixes are contaminated

IsoPol BST⁺ polymerase has high tolerance to matrix effects. LAMP was performed using λ DNA template in presence of human serum demonstrating that IsoPol BST⁺ maintains high activity over a wide range of serum concentrations. No amplification was detected in the absence of template.

Limited sample

Small sample size and low sample concentration are often limitations in molecular diagnostic testing. In addition to enhanced inhibitor tolerance, the high quality and purity of IsoPol polymerases make them ideal for amplification of most sample types, especially small and impure samples.



IsoPol® BST⁺ : Glycerol FREE "Lyophilisation and automation friendly"

One of the major challenges that manufacturing companies face is the handling of kit components, such as enzymes or proteins, during process optimization and shipping. When handled incorrectly they can aggregate, denature, degrade, and lose activity. A general practice to improve user experience and reduce cold chain shipping costs is lyophilisation.

For easier adaptation to lyophilisation, the IsoPol BST⁺ is also offered in a Glycerol FREE version.

Furthermore, in compliance with the EU REACH regulation, the IsoPol BST⁺ formulations are offered Triton FREE.

IsoPol BST⁺ High Concentration Glycerol FREE

IsoPol BST⁺ High Concentration Glycerol FREE (IsoPol BST⁺ HC Glycerol FREE) is used to ease the product development processes in sequencing technologies, solid phase amplification and several isothermal technologies.

Same performance as IsoPol BST⁺ (i.e., high processivity, relative SDA, and relative activity) with the only difference that it is offered in a 2X storage buffer at a concentration > 500 U/µl.

IsoPol BST⁺ Glycerol FREE

IsoPol BST⁺ Glycerol FREE is the popular choice for isothermal applications such as LAMP and RT-LAMP at point-of-care diagnostics for its superior amplification performance and robustness. The enzyme is offered in a 1X storage buffer at a concentration < 300 U/µl.

IsoPol® selection guide

	Temperature range	Optimal temperature	Strand displacement with salt	Salt tolerance (NaCl/KCL) (mM)	Specific activity (U/mg)	Applications
IsoPol® BST⁺	25 - 65°C	65°C	++++	50-350	40,000	LAMP, RT-LAMP, RAM,NEMA, HDA, MDA/SDA, RCA, RPA
IsoPol® BST⁺ HC Glycerol FREE	25 - 65°C	65°C	++++	50-350	40,000	Sequencing technologies, solid phase amplification
IsoPol® SD⁺	20 - 42°C	37°C	++++	100-350	10,000	MDA/SDA, RPA, HAD
IsoPol® DNA Polymerase	20 - 42°C	37°C	++	25-110	15,000	MDA/SDA, RPA, HAD

++++ Optimal recommendation for selected application(s)

++ Works well for selected application(s)

Abbreviations

MDA / SDA	Multiple / Strand displacement amplification
RCA	Rolling circle amplification
RPA	Recombinase Polymerase amplification
SMART	Signal mediated amplificaiton of RNA technology
LAMP	Loop mediated isothermal amplificaiton
RAM	Ramification amplification
HDA	Helicase-dependent amplification
NASBA	Nucleic acid sequence-based amplificaiton
NEMA	Nicking enyzme-mediated amplification

IsoPol DNA polymerases are compatible with a wide range of buffer formulations and isothermal applications. Please refer to the IsoPol selection guide above for guidance on which polymerase is more suitable for your use or get in touch with us at contact@arcticzymes.com.

No license required

At ArcticZymes, we pride ourselves on always offering seamless accessibility to our high quality products. Produced under ISO 13485, our enzymes are sold under a “no license required” policy to ensure that our customers are not restricted by legal burdens, now or with their future use. In addition, we offer our nucleases in a flexible format and are readily available to discuss your custom needs.

Ordering information

	Article no.	Pack size*	Concentration
IsoPol® BST⁺ HC Glycerol FREE**	71522-100	Custom	> 500 U/µl
IsoPol® BST⁺ Glycerol FREE	71512-100	Custom	< 300 U/µl
IsoPol® BST⁺	71502-201	200 U	5 U/µl
	71502-100	Custom	Custom
IsoPol® BST⁺ 10X Reaction Buffer Pack	71502-001	4 x 1,5 ml	10X
IsoPol® SD⁺	71501-201	200 U	5 U/µl
	71501-100	Custom	Custom
IsoPol®	71500-201	200 U	5 U/µl

* One unit is defined as the amount of enzyme that will incorporate 10 nmol of dNTP into acid insoluble material in 30 minutes at 37°C for IsoPol DNA Polymerase and IsoPol SD⁺ and at 65°C for IsoPol BST⁺.

** IsoPol BST⁺ HC Glycerol FREE is provided in a 2X storage buffer. For more information, please contact support@arcticzymes.com.

Your OEM Partner to deliver novel solutions for genomics and proteomics

Quality

ArcticZymes is dedicated to the quality of its products and is certified according to ISO 13485:2016. ArcticZymes offers the convenience of providing standard bulk enzymes as off the shelf products. In addition, ArcticZymes offers IsoPol enzymes in customized formats.

Additional information

We are pleased to provide further information relating to IsoPol enzymes such as ssDNA / dsDNA endonuclease and exonuclease activity, purity, $Mg2^{+}$, pH, processivity, activity and strand displacement data.

For more information, please check our website www.arcticzymes.com or contact us.

ArcticZymes Technologies

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