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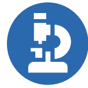 Laborgeräte & Service

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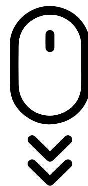
See the following pages for more information!



Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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HotStart SD Polymerase

Catalog No. SD-28-1000, 1000 U

DESCRIPTION

HotStart SD polymerase is a variation of Taq polymerase (blocked by Taq antibody) with strong strand displacement activity and processivity. Two key changes turn ordinary Taq polymerase into a polymerase that handles the most Stringent Demands (SD): high GC content, strong secondary structure, long distance amplification, and the latest amplification assays. It often provides greater final product yields.

Unlike natural enzymes with strong strand displacement activity such as Phi29 or Bst polymerase, which are active only below 68 °C, SD polymerase is stable up to 93 °C. SD performs isothermal amplifications like LAMP; however, now you can add an initial denaturation step (92 °C) or hot-start for increased specificity. SD polymerase can perform long-distance PCR with extremely low concentrations of template. SD polymerase is particularly effective for PCDR, a new PCR assay that incorporates strand displacement, particularly useful in creating more sensitive qPCRs.

ACTIVITIES

HotStart SD polymerase has 5’-3’ polymerase and 5’-3’ strand displacement activities. It does not have any exo-/endonuclease activity. The enzyme does A-overhangs. HotStart efficiency tested.

THERMOSTABILITY

SD polymerase is thermostable up to 93 °C but not as stable as regular Taq polymerases. In a denaturation step with LAMP or PCR amplification, 92 °C or even lower that 92 °C is strongly recommended. Please find a standard program suggested in this text under ″PCDR″.

CONCENTRATION

10 units/µl

STORAGE BUFFER

10 mM K-phosphate buffer pH 7.0, 100 mM NaCl, 0.5 mM EDTA, 1 mM DTT, 0.01% Tween 20; 50% glycerol (v/v)

10X REACTION BUFFER

pH8.8, with Tween-20 and BSA added; add $MgCl_2$ to a final concentration of **3-3.5 mM** before use.

STORAGE TEMPERATURE

Store at -20°C in a constant temperature freezer

REFERENCES

1. Claire L, et al. Polymerase chain displacement reaction. Biotechniques. 54: 93-97, 2013
2. Ignatov KB, et al. A strong strand displacement activity of thermostable DNA polymerase markedly improves the results of DNA amplification. Biotechniques. 57: 81-87, 2014

APPLICATIONS:

- ***LAMP (Loop-mediated isothermal amplification):*** The optimum temperature range for LAMP amplification is 62–68 °C. The high thermostability of the enzyme allows carrying out LAMP with an initial DNA denaturation step (92 °C for 2 min), which will enhance the reaction in some applications. LAMP can be performed with 15 – 50 units of SD Polymerase per 50 µl.
- ***PCR:*** SD Polymerase is suitable for amplification of short (from 100 bp) and long (up to 20 – 30 kb) DNA fragments from simple (plasmids) and complex (genomic DNA) templates. In PCR applications SD Polymerase demonstrates higher yields, speed and efficiency compared with Taq. Even single copy templates are amplified with good results. We recommend 92 °C for denaturation steps, 68 °C for elongation steps in PCR and 0.5-2 units of SD Polymerase per 50 µl for PCR.
- ***PCDR (Polymerase Chain Displacement Reaction):*** SD polymerase is the enzyme of choice to run PCDR, providing much higher efficiency than Bst. Here the enzyme has outstanding performance due to its unique combination of strand displacement and thermostability. We recommend 92 °C for denaturation steps, 68 °C for elongation steps and 0.5 – 2 units of SD Polymerase per 50 µl for PCDR.
- **Example for usage in PCR or PCDR:** The 50 µl reaction mixture includes 10 units of SD Polymerase, 1x SD Polymerase Reaction buffer; 3 mM $MgCl_2$; 0.375 mM dNTPs (each); 20 pmol of each inner primer, 10 pmol other primers (each); about 0.05 ng of cDNA library as a template. PCR or PCDR thermocycler program: Preheating: 92°C, 1 min, 25 cycles (92 °C, 30 sec; 60 °C, 30 sec; 68 °C, 30 sec).

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