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ID*PROOF*[™] DNA Polymerase

Cat #

IDL008 500U

Concentration

1 ul contains 5 units

Description

 ${
m ID}PROOF^{
m TM}$ is a high performance complex of enzyme with additives. It is used to improve the reliability and yield of conventional primer extension reaction. ${
m ID}PROOF^{
m TM}$ has two advantages: (1) high fidelity with an error frequency $1.6 / 10^6$ (or $0.0016 / 10^3$) during DNA synthesis and (2) ${
m ID}PROOF^{
m TM}$ increases the efficiency of polymerization reaction, resulting in a great percentage of extenuation reaction completion up to 10 kb to 30 kb. Optimum temperature is between 72-78° C and remains > 95% active following 1-hour incubation at 95° C.

10X Reaction Buffer

200 mM TrisHCl (pH 8.8)

100 mM KCl

100 mM (NH₄)₂ SO₄

20 mM Mg SO₄

1% Triton X-100

1 mg / ml bovine serum albumin (BSA)

Reaction Conditions

Note: All reagents, including IDPROOFTM, should be mixed immediately before use.

DNA synthesis is performed in $100\mu l$ of mixture containing $20\text{-}200\mu M$ dNTPs, $0.3\text{-}1~\mu M$ Primers, 0.1-0.250 ng of template DNA, $10~\mu l$ of 10~x reaction buffer and 2.5-5 units of $IDPROOF^{TM}$. Mix the reaction gently, centrifuge briefly and then overlay with light mineral oil. Initially, denature the reaction by incubating at 95° C for 5 minutes and then cool to $40\text{-}68^{\circ}$ C for 5 minutes to allow the primers to anneal to the template DNA.

It is important to add the reaction components in the following order:

- 1- H₂O
- 2. 10x reaction buffer
- 3-dNTPs
- 4- DNA template and primers
- 5- IDPROOFTM

References

- (1) Kaledin, A.S., et al (1980) Biokhimiya, 45, 494
- (2) Kaledin, A.S., et al (1981) Biokhimiya, 46, 1576
- (3) Kaledin, A.S., et al (1982) Biokhimiya, 47, 1785
- (4) Ruttiman, C., et al (1985) Eur J Biochem, 149, 41
- (5) Varghese, R., et al (1998) Endocrinology, 139 (11), 4714
- (6) Osiowy, C., et al (1998) Microbiology, 36 (11), 3149
- (7) Ying-Chi, IP., et al (2000) Endocrinology, 141 (4), 1356
- (8) Nadler, S., et al (2000) J. Parasitol., 86 (3), 588
- (9) Crack, J., et al (2002) J. Biochem., 269, 933
- (10) Pierce, S. K., et al (2007) Symbiosis, 43, 57

Storage

-20° C in a constant temperature freezer. Stable for at least one year.

Do not freeze-thaw multiple times.

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