ProofDog HiFi DNA Polymerase

Products

CAT. #	Description	Volume
PFDG001S	ProofDog HiFi DNA Polymerase with 5x HF Buffer (no dNTP), (25 Units)	25 Units, 5 µl
PFDG010	ProofDog HiFi DNA Polymerase with 5x HF Buffer (no dNTP), (100 Units)	100 Units, 20 µl
PFDG050	ProofDog DNA Polymerase with 5x HF Buffer (no dNTP), (500 Units)	500 Units, 5 x 20 µl

Product Information

Bulldog Bio's Proofdog HiFi DNA Polymerase is a novel recombinant polymerase that contains a 5' → 3' DNA polymerase and a 3 '→ 5' exonuclease domains along with a Sso7D tethering domain. The ProofDog HiFi DNA polymerase achieves 55-fold higher fidelity than TAQ DNA polymerase and generates blunt end PCR products. The ProofDog HiFi DNA polymerase can generate DNA fragments up to 10 Kbp in length with higher accuracy and two-to-three-fold greater processivity than TAQ DNA polymerase.
Extension: 10,000 base-pair extension has been confirmed
Speed: 3 Kbp / minute

ProofDog HiFi DNA polymerase has 5' – 3' polymerase activity, 3' – 5' exonuclease activity, and produces amplicons with blunt ends.

Product Components

	500 U	100 U	25 U
ProofDog HiFi DNA Polymerase	5 x 20 µl	1 x 20 µl	1 x 5 µl
5X HF Reaction Buffer	20 x 1.5 ml	4 x 1.5 ml	1 x 1.5 ml
GC Extender	5 x 1.5 ml	1 x 1.5 ml	1 x 1.5 ml

All components are to be stored at - 20 °C

Protocol

1. Add following components to a PCR tube on ice:

Note: DNA amount, primer amount, ProofDog HiFi Polymerase units and $MgSO_4$ concentration should be optimized.

Component	Volume	Final Concentration
Template DNA (up to 500 ng)	up to 10 µl	–
10 µM Forward Primer	0.5 µl – 2.5 µl	100 nM – 500 nM
10 µM Reverse Primer	0.5 µl – 2.5 µl	100 nM – 500 nM
5X HF Buffer	10 µl	1 X
10 mM dNTP mix	1 µl	0.2 mM
ProofDog HiFi DNA Polymerase	0.5 µl – 1µl	2.5 U – 5 U
PCR Enhancer (Optional)*	5 µl – 15 µl	–
Nuclease-free H_2O	up to 50 µl	–

*PCR Enhancer is intended for difficult-to-amplify templates (e.g., GC content > 60 % or for templates longer than 2.5 Kbp). Volume of PCR Enhancer required (5 µl – 15 µl) should be optimized for each template and primer combination.

2. Mix contents (either through pipetting multiple times or briefly vortexing).

3. Centrifuge briefly and proceed to PCR amplification steps.

4. PCR amplification:

Note: Number of cycles, annealing temperature and extension should be optimized for each template and primer set.

Stage	Step	Temperature (°C)	Time	Cycles
1	Denature*	98	5 sec - 30 sec	20-30
	Anneal	55 - 60	5 sec - 30 sec	
	Extend	72	1 Kbp / 20 sec	
2	Final extension step	72	5 min	1
3	Holding step	4	∞	1

5. Product is ready for downstream analysis. For long-term storage keep the PCR reactions at -20C.

END OF PROTOCOL