

## TruFidelis Core DNA Polymerase

FB02008010 100 Units

FB02008050 500 Units

Store at -25°C to -15°C

TruFidelis Core DNA Polymerase is a high-fidelity, proofreading DNA polymerase combining a novel enzyme with a processivity-enhancing domain. It offers high performance for all major PCR applications. It generates long amplicons with accuracy and speed, even on challenging templates. The high fidelity makes the TruFidelis Core DNA Polymerase a superior choice for cloning.

- > 50X fidelity compared to Taq polymerase.
- 5'→3' DNA polymerase activity.
- 3'→5' exonuclease activity.
- Generates blunt-end products.
- Amplifies long amplicons such as 7.5 kb genomic and 20 kb  $\lambda$  DNA.

### Kit Contents

Reagents	100 Units	500 Units	Description
TruFidelis Core DNA Polymerase	50 $\mu$ L	250 $\mu$ L	-
5X TruFidelis Core HF Buffer	2 x 1.5 mL	6 x 1.5 mL	Provides 1.5 mM MgCl <sub>2</sub> at 1X concentration. Recommended as default buffer.
5X TruFidelis Core GC Buffer	1.5 mL	2 x 1.5 mL	Provides 1.5 mM MgCl <sub>2</sub> at 1X concentration. Recommended for GC-rich amplicons
50 mM MgCl <sub>2</sub> solution	1.5 mL	2 x 1.5 mL	For increasing Mg <sup>2+</sup> final concentration, if desired.
DMSO	500 $\mu$ L	500 $\mu$ L	Recommended for GC-rich amplicons. DMSO is not recommended for amplicons with very low GC % or amplicons that are > 20 kb.

### PCR Reaction Setup:

For multiple reactions, prepare a master mix of components common to all reactions to minimize pipetting error, then dispense appropriate volumes into individual PCR tube before adding template DNA.

**Note:** It is critical that the TruFidelis Core DNA Polymerase is the last component added to the PCR mixture, since the enzyme exhibits 3'→5' exonuclease activity that can degrade primers in the absence of dNTPs.

Component	20 $\mu$ L Reaction	50 $\mu$ L Reaction	Final Concentration
Water, nuclease-free	to 20 $\mu$ L	to 50 $\mu$ L	-
5X TruFidelis Core HF Buffer	4 $\mu$ L	10 $\mu$ L	1X
10 mM dNTP Mix	0.4 $\mu$ L	1 $\mu$ L	0.2 mM each
Forward Primer	varies	varies	0.5 $\mu$ M*
Reverse Primer	varies	varies	0.5 $\mu$ M*
Template DNA	varies	varies	50-250 ng (gDNA) or 1 pg-10 ng (plasmid DNA) per 50 $\mu$ L reaction
DMSO, optional	0.6 $\mu$ L	1.5 $\mu$ L	3%
TruFidelis Core DNA Polymerase	0.2 $\mu$ L	0.5 $\mu$ L	0.02 U/ $\mu$ L**

\*The recommendation for final primer concentration is 0.5  $\mu$ M, but it can be varied in a range of 0.2-1.0  $\mu$ M, if needed.

\*\*Optimal amount of enzyme can range from 0.5 to 2 units per 50  $\mu$ L reaction depending on amplicon length and difficulty. It is not recommended to exceed 2 U/50  $\mu$ L (0.04 U/ $\mu$ L), especially for amplicons that are > 5kb.

### Thermal Cycling Conditions on Thermal Cycler:

Step		2-step protocol (for primers with T <sub>m</sub> values ≥ 69°C)		3-step protocol	
		Temp.	Time	Temp.	Time
Initial denaturation		98°C	30 sec	98°C	30 sec
25-35 PCR cycles	Denature	98°C	5-10 sec	98°C	5-10 sec
	Anneal	72°C	15-30 sec/kb	varies	10-30 sec
	Extend			72°C	15-30 sec/kb
Final extension		72°C	5-10 min	72°C	5-10 min
		4°C	hold	4°C	hold

Use your PCR product immediately in downstream applications, or store it at -20°C.