TruFidelis Plus Green PCR Master Mix

FB02007010 100 Reactions
FB02007050 500 Reactions
Store at -25°C to -15°C

TruFidelis Plus Green PCR Master Mix is a ready-to-use mixture of DNA polymerase, salts, magnesium, and dNTPs for efficient PCR amplification. The master mix is ideal for applications where accuracy is important (cloning, sequencing, site directed mutagenesis). Two tracking dyes and a density reagent are pre-added to the master mix for direct loading of PCR products onto gels.

The master mix contains TruFidelis Plus DNA Polymerase, a high-fidelity, proofreading DNA polymerase combining a novel enzyme with a processivity-enhancing domain. It features a hot start mechanism, allowing room temperature reaction setup and storage of pre-assembled PCR reactions.

- 5′→3′ DNA polymerase activity.
- 3′→5′ exonuclease activity.
- Generates blunt-end amplification products.
- Amplifies up to 10 kb from genomic DNA and up to 20 kb from low complexity DNA.
- > 100X fidelity compared to Taq polymerase.

Kit Contents

Reagents	100 Reactions	500 Reactions	Description
2X TruFidelis Plus Green PCR Master Mix	2 x 1.25mL	10 x 1.25 mL	Provides 1.7 mM $MgCl_2$ at 1X concentration
5X TruFidelis Plus GC Enhancer	1.25 mL	4 x 1.25 mL	Recommended for targets with >65% GC Sequence
Water, nuclease-free	2 x 1.25 mL	10 x 1.25 mL	-

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.

Component	20 µL Reaction	50 µL Reaction	Final Concentration
Water, nuclease-free	to 20 µL	to 50 µL	-
2X TruFidelis Plus Green PCR Master Mix	10 µL	25 µL	1X
Forward Primer	varies	varies	0.5 µM*
Reverse Primer	varies	varies	0.5 µM*
Template DNA	varies	varies	5-100 ng (gDNA) or 0.01-10 ng (plasmid DNA) per 50 µL reaction
5X TruFidelis Plus GC Enhancer (optional)	4 µL	10 µL	1X

*Reduce the final primer concentration to 0.2 µM when amplifying >5 kb targets from high complexity DNA and multiplex reactions.

Thermal Cycling Conditions on Thermal Cycler:

Step		2-step protocol (for primers >30 nt in length)		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 sec
	Extend			72°C	15-30 sec/kb
Final extension		72°C	5 min	72°C	5 min
		4°C	hold	4°C	hold

Load the PCR mixture directly on a gel.