

i7 High-Fidelity DNA Polymerase 2x Master Mix

Catalog #	3257	3259
Package Size	100 Reactions	500 Reactions

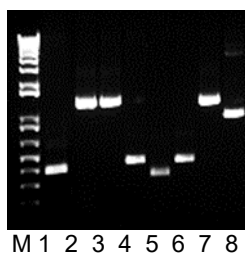
Description

Intact Genomics (IG) i7 high-fidelity DNA Polymerase 2x master mix is ready to use premix which contains i7 high-fidelity DNA Polymerase, dNTPs, MgCl₂, PCR enhancers and stabilizers with optimized proprietary reaction buffer. i7 high-fidelity DNA Polymerase is a genetically engineered, heat stable DNA polymerase which has 5'→3' polymerase and 3'→5' exonuclease (proofreading) activities. This enzyme has high-fidelity, sensitivity and processivity with an error rate ~2.8x10²-fold lower than *Taq* DNA polymerase, and significantly lower than other proofreading enzymes in the marketplace (1). This 2x master mix product is supplied with 5x magic enhancer that enables efficient amplification of GC rich templates up to 84%.

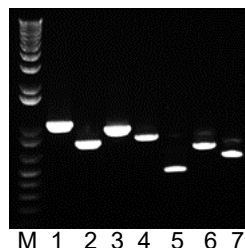
Activity Data

We have tested i7 high-fidelity 2x master mix with difficult templates for PCR amplification. Typical PCR results are shown below:

A). Colony PCR



B). PCR to detect difficult templates



Applications

- Long and difficult template DNA amplification
- Cloning
- High-fidelity PCR
- Efficient for amplifying high GC content template DNA with magic enhancer.

Product Includes

- i7 high-fidelity DNA Polymerase 2x master mix
- 5x Magic Enhancer

Storage Temperature

-20°C

Heat Inactivation

No

Storage Buffer

50 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, pH 7.5 @ 25°C

Protocol

1. Thaw i7 high-fidelity 2x master mix, primer solutions, 5x magic enhancer (if required) and mix thoroughly before use.
2. Prepare a reaction mix according to the following table:
(The reaction mix typically contains all the components needed for PCR except the template DNA).

Components	20 µl reaction	50 µl reaction	Final concentration
Template DNA	variable	variable	1-1000 ng
Forward primer (10 µM)	1.0 µl	2.5 µl	0.5 µM
Reverse primer (10 µM)	1.0 µl	2.5 µl	0.5 µM
5x magic enhancer (optional)	(4.0 µl)	(10.0 µl)	(1x)
i7 high-fidelity 2x Master Mix	10.0 µl	25.0 µl	1x
H ₂ O up to	20.0 µl	50.0 µl	

3. Mix the reaction mixture thoroughly.
4. Add template DNA to the individual PCR tube containing the reaction mixture.
5. Program the thermal cycler according to the manufacturer's instructions. A typical PCR cycling program is outlined in the following table.

PCR Cycling Conditions:			
Steps	Temp.	Time	Cycles
Initial denaturation	98 °C	1-2 min	1
Denaturation	98 °C	10-20 sec	25-35
Annealing	52-66 °C	10-30 sec	
Extension	68-72 °C	10-30 sec/kb	
Final extension	68-72 °C	5 min	1
Hold	4-12 °C	∞	

References

1. Frey, B. and Suppmann, B. (1995). *BioChemica*. 2, 34-35.

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5. Program the thermal cycler according to the manufacturer's instructions. A typical PCR cycling program is outlined in the following table.

PCR Cycling Conditions:			
Steps	Temp.	Time	Cycles
Initial denaturation	98 °C	1-2 min	1
Denaturation	98 °C	10-20 sec	25-35
Annealing	54-66 °C	10-30 sec	
Extension	68-72 °C	10-30 sec/kb	
Final extension	68-72 °C	5 min	1
Hold	4-12 °C	∞	

6. Place the PCR tubes in the thermal cycler and start the cycling program.