Life Technologies™
Your Molecular & Cell Technology Partner

i7® High-Fidelity DNA Polymerase 2x Master Mix

Intact Genomics®

Catalog #	3257	3259
Package Size	100 Reactions	500 Reactions

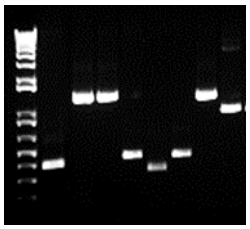
Description

Intact Genomics (ig®) i7® High-Fidelity DNA Polymerase 2x Master Mix is ready to use premix which contains i7® high-fidelity DNA Polymerase, dNTPs, $MgCl_2$, 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). Proprietary buffer allows for amplification of non GC rich templates and of GC rich templates up to 84%.

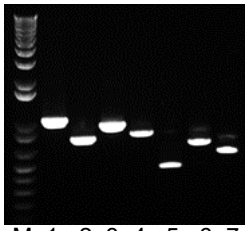
Activity Data

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

Product Includes

- i7® High-Fidelity DNA Polymerase 2x Master Mix

Storage Temperature

-20ºC

Heat Inactivation

No

Storage Buffer

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

Protocol

1. Thaw i7® high-fidelity 2x master mix and primer solutions 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
i7® high-fidelity 2x Master Mix	10.0 µl	25.0 µl	1x
H_2O 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.
3. Program the thermal cycler according to the manufacturer's instructions. A typical PCR cycling program is outlined in the following table:
6. Place the PCR tubes in the thermal cycler and start the cycling program.

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

References

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

Related Products

- Taq DNA Polymerase 2x Premix (Cat.# 3249)
- ig® 10B Electrocompetent Cells (Cat.# 1212-12)
- ig® 10B Chemically Competent Cells (Cat.# 1011-12)
- ig-Fusion™ Cloning Kit (Cat.# 4111)
- i7® Hot Start High-Fidelity DNA Polymerase (Cat.#3281)
- i7® High-Fidelity DNA Polymerase (Cat.#3254)

Technical Support

Intact Genomics is committed to supporting the worldwide scientific research community by supplying the highest quality reagents. Each new lot of our products is tested to ensure they meet the quality standards and specifications designated for the product.
Please follow the instructions carefully and contact us if additional assistance is needed. We appreciate your business and your feedback regarding the performance of our products in your applications.

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