



i7® High-Fidelity DNA Polymerase

Manual

Catalog #	3254	3255	
Package Size	200 units	500 units	
Concentration	2 units/μl		



Important!

-20°C Storage Required

- * Immediately inspect packages
- * Freeze upon receipt

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i7[®] High-Fidelity DNA Polymerase

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Description:

Intact Genomics (ig®) i7® High-Fidelity DNA Polymerase is a genetically engineered, heat stable DNA polymerase which has $5' \rightarrow 3'$ polymerase and $3' \rightarrow 5'$ exonuclease (proofreading) activities. This enzyme has the high-fidelity, sensitivity and processivity with an error rate ~2.8x10² -fold lower than Taq DNA polymerase, and significantly lower than the error rates of other proofreading enzymes in the marketplace (1). i7® High-Fidelity DNA Polymerase is ideal for cloning and can be used for long (up to 20kb) or difficult amplicons. This product is supplied with the Intact Genomics proprietary 2.5 x i7® PCR reaction buffer containing MgCl₂ with a final (1x) concentration of 2 mM. This buffer allows for amplification of non GC rich templates up to 84%.

Protein Purity:

The physical purity of this enzyme is ≥98% as assessed by SDS-PAGE with Coomassie® blue staining (see figure 1).

Product Source:

E. coli strain expressing genetically engineered i7® High-Fidelity DNA Polymerase gene.

KDa 1 2 200 116 97 66 45 30

Figure 1: Lane 1. Protein marker Lane 2. i7 High-Fidelity DNA Polymerase

Applications:

- Long and difficult template DNA amplification
- Cloning
- High-fidelity PCR

Product Components:

- i7[®] High-Fidelity DNA Polymerase
- 2.5x i7® PCR Buffer with Mg2+

Storage Buffer:

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



Storage Temperature:

-20ºC

Heat Inactivation:

No

Quality Control Assays:

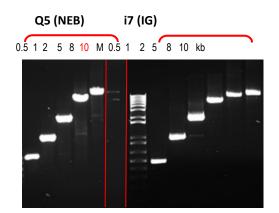
i7[®] High-Fidelity DNA Polymerase is free from detectable nuclease activities.

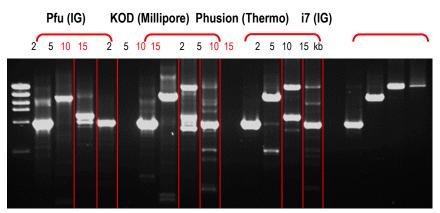
Unit Definition:

One unit is defined as the amount of enzyme that incorporates 10 nmoles of dNTP into acid-insoluble form in 30 minutes at 72° C.

Comparison Data:

We have tested i7® High-Fidelity DNA Polymerase activity with λ DNA and other difficult templates for PCR amplification up to 20kb. Intact Genomics (IG) i7 High-Fidelity DNA Polymerase generates robust and high-quality PCR products in comparison with other high-fidelity DNA polymerases available in the marketplace (data shown below):







Protocol:

- 1. Thaw 2.5x i7® PCR Buffer, dNTP mix, 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.)

PCR Reaction Set Up:			
Template DNA	x μl (0.01-0.5 μg)		
2.5 x i7 PCR Buffer	20.0 µl		
dNTP (10 mM)	1.0 µl		
Forward Primer (10 µM)	2.5 µl		
Reverse Primer (10 µM)	2.5 µl		
i7 High-Fidelity	0.5 μΙ		
DNA Polymerase (2 U/μl)			
H₂O up to	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 ℃	1-2 min	1		
Denaturation	98 °C	10-20 sec			
Annealing	54-66 °C	10-30 sec	25-35		
Extension	68-72 °C	10-30 sec/kb	20-00		
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.



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 2X Master Mix (Cat.# 3257)

Ordering Information:

- Order online within the USA. Place orders on www.intactgenomics.com using our secure Shopping Cart.
- Order by email, phone, or fax.

Email: sales@intactgenomics.com

Phone: (314) 942-3655 | Toll-free: 855-835-7172 | Fax: (314) 942-3656

• Order via our distributors.



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Our hours are Monday - Friday, 8AM to 5PM, U.S. Central Standard Time.

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