proceeding the second			
Intact Genomics®	Pfu 2x	Master	Mix

Catalog #	3325	3326	
Package Size	50 reactions	200 reactions	
Volume	1.25 ml	5 ml	
Concentration	N/A		

## Description

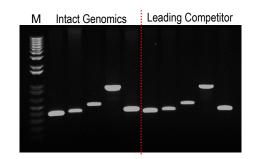
*Pfu* DNA Polymerase 2x master mix is ready to use premix which contains *Pfu* DNA Polymerase, dNTPs, MgCl<sub>2</sub> and stabilizers with optimized reaction buffer. It has been optimized for routine PCR applications. *Pfu* DNA polymerase is a heat stable DNA polymerase which has  $5' \rightarrow$ 3' DNA polymerase and  $3' \rightarrow 5'$  exonuclease (proofreading) activities. *Pfu* DNA polymerase retains the high fidelity, sensitivity and processivity with an error rate six-fold lower than *Taq* DNA polymerase, and significantly lower than the error rates of most other proofreading enzymes or DNA polymerase mixtures (1). This product is supplied with the unique Intact Genomics 5x Magic Enhancer that enables efficient amplification of GC rich templates up to 84%.

#### **Product Source**

E. coli strain expressing a *Pfu* DNA Polymerase gene from Pyrococcus furiosus.

# Pfu DNA Polymerase Comparison Data

We repeatedly compare our *Pfu* Polymerase side-by-side with a leading competitor for PCR assay. Our enzyme is better than the competitor. A typical gel picture is shown below:



## Applications

- Routine PCR
- Primer extension
- Colony PCR
- Genotyping
- Efficient for amplifying high GC content template DNA
  with Magic Enhancer

#### **Product Includes**

- 1) Pfu 2x master mix
- 2) 5x Magic Enhancer

# Storage Temperature

-20 °C

## **1x Master Mix Composition**

10 mM Tris-HCl pH 9.0 50 mM KCl 1.5 mM MgCl<sub>2</sub> 0.2 mM dNTPs 5% Glycerol 0.08% Igepal CA 630 0.05% Tween-20 100 Units/ml Pfu DNA Polymerase.

# Protocol

1. Prepare a reaction mix according to the following table:

PCR reaction set up:		
Template DNA	1-50 ng	
Forward primer (5 µM)	1.0 µl	
Reverse primer (5 µM)	1.0 µl	
Pfu 2x master mix	10.0 µl	
5x Magic Enhancer (optional)	4.0 µl	
H <sub>2</sub> O up to	20.0 µl	

- 2. Mix the reaction mixture thoroughly.
- 3. 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	95 °C	3 min	1		
Denaturation	95 °C	30 sec			
Annealing	50-66 °C	30 sec	25.40		
Extension	72 °C	1 min/kb	25-40		
Final Extension	72 °C	5 min	1		
Hold	4-12 ⁰C	∞			

- 5. Place the PCR tubes in the thermal cycler and start the cycling program.
- Analyze 5 µl of PCR products by agarose gel electrophoresis.

## Reference

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