Product code: 2MMCP- Lot Number:



# MegaMix Crystal Q5 High Fidelity Polymerase

P. Code	Size in 100 Rxn	Size in 500 Rxn	Component	Description	Lot Number	Expiry
2MMCB-1	1 x 1 mL	5 x 1 mL	5X MegaMix Crystal Q5 HiFi Reaction Buffer	Specially optimised reaction buffer containing dNTPs, $MgSO_4$ and enhancers.		
2MMCE-0.05	1 x 50 μL	5 x 50 μL	MegaMix Crystal Q5 HiFi DNA Polymerase, 2 U/μL	Q5 Hot Start High Fidelity Polymerase in storage buffer.		
MFO-1	1 x 1 mL	5 x 1 mL	4X microFORCE	Buffer solution optimised for templates with high GC content or GC-repeats.		
5JWA-1	3 x 1 mL	15 x 1 mL	Just Water (Molecular Grade Water)	Aliquoted, Quality controlled, nuclease-free, molecular grade water.		

## **Applications**

- Long-range PCR amplifications (Up to 40 kb)
- NGS library preparation and Sequencing
- Site-directed Mutagenesis
- Amplification of difficult (GC-rich) templates

## **Product Description**

### 5X MegaMix Crystal HiFi Reaction Buffer

Special reaction buffer to be used with MegaMix Crystal Q5 High Fidelity DNA Polymerase, which includes optimal  $Mg^{2+}$  concentration and dNTP amount, and proprietary formulation with stabilisers and enhancers specifically optimised for our enzyme.

## MegaMix Crystal Q5 High Fidelity DNA Polymerase 2 U/μL

The ideal choice for complex PCR applications where the highest fidelity is required. MegaMix Crystal Q5 High Fidelity DNA Polymerase provides a 280-fold higher fidelity compared to Taq Polymerase and a 5-fold higher fidelity compared to standard Pfu Polymerases. Moreover, the enzyme is Hot-Start for increased specificity and sensitivity, also allowing room temperature reaction setup.

It is the ideal enzyme for cloning and various sequencing applications, including NGS and sanger. The PCR products produced are blunt-ended and can be ligated into blunt vectors.

## 4X microFORCE

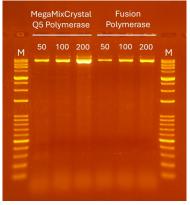
Microzone's enhancing buffer for the amplification of difficult templates.

#### Just Water

Our molecular biology grade water, providing high purity with the complete absence of DNase and RNase.

## **Key Features**

- Highest fidelity amplification (~280X higher than Taq)
- Suitable for amplicons up to 40 kb
- Ultra-low error rate due to 3´-5´ exonuclease proofreading activity
- Ideal for cloning, sequencing and long or difficult amplicons
- Hot-Start version for increased specificity and sensitivity



Superior amplicon yield using MegaMix Crystal Q5 High Fidelity Polymerase vs commercially available Fusion polymerases. Gel image shows amplification of 8.5 kb fragment from human gDNA, using template concentrations of 50, 100 and 200 ng. PCR was performed in 50 μL reaction using Microzone's MegaMix Crystal Q5 High-Fidelity range. M: 1 kb DNA Ladder.

## Protocol

Thaw all reagents completely and mix well before use.

Components	Volume	
5X MegaMix Crystal HiFi Reaction Buffer	10 μL	
MegaMix Crystal Q5 High Fidelity DNA Polymerase, $2U/\mu L$	0.5 μL	
4X microFORCE (optional: for GC-rich templates only)	12.5 μL	
Primers	xμL	
Template	γ μL	
Just Water (Molecular Grade Water)	make up to 50 μL	

Prepare a master mix as described in the table below.

Mix gently, avoiding bubbles, centrifuge if necessary.

Include a no template control and positive control as required.

# Thermocycling

The following general cycling conditions are intended for use as a guide and can vary depending on the template and primers being used.

For the amplification of long fragments contact Microzone for recommendations.

Step	Cycles	Temperature	Time
Initial Denaturation	1	98°C	30 seconds
Denaturation		98°C	5-10 seconds
Annealing	25 - 35	60 - 68°C	10-15 seconds
Elongation		72°C	30 seconds / kb
Final Elongation	1	72°C	2 min

For Research Use Only. Not for use in diagnostic procedures.

### Product Handling and Storage

To ensure the quality of the product until the expiry date keep at the recommended storage temperature and limit exposure to light.

### Licenses

This product is covered by one or more patents.

This product is licensed for research and commercial use from Bio-Rad Laboratories, Inc., under U.S. Pat. Nos. 6,627,424, 7,541,170, 7,670,808, 7,666,645, and corresponding patents in other countries. No rights are granted for use of the product for Digital PCR or real-time PCR applications, with the exception of quantification in Next Generation Sequencing workflows.

Simple | Effective | Efficient