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# GemTag<sup>™</sup> Hot Start 5xMaster Mix

(Catalog #EPM31, #EPM32, #EPM33, #EPM34)

## FOR LABORATORY USE ONLY

## **Description**

GemTaq<sup>TM</sup> Hot Start 5xMaster Mix was developed for ultimate convenience, time conservation and greater productivity. It is a ready-to-use 5x PCR reaction mix containing GemTaq<sup>TM</sup> Hot Start DNA Polymerase, dNTPs, MgCl<sub>2</sub> and reaction buffer at optimal concentrations for efficient amplification of DNA templates. Users of the GemTaq<sup>TM</sup> Hot Start 5xMaster Mix only need to add water, primers and template to perform PCR assays.

Use of the GemTaq<sup>TM</sup> Hot Start 5xMaster Mix provides robust yields and high sensitivity of PCR amplifications within a short reaction time. The superior performance of this mix is due to properties of GemTaq<sup>TM</sup> Hot Start DNA Polymerase that is a heat-activated high-performance proprietary formulation of GemTaq<sup>TM</sup> DNA Polymerase. The modified polymerase activity is restored during the initial denaturation step (94-95 °C for 3 min.) allowing for a hot-start PCR. Therefore GemTaq<sup>TM</sup> Hot Start DNA Polymerase provides improved specificity and very high PCR sensitivity by eliminating the presence of non-specifics products.

## **Advantages**

- Ultimate convenience, time conservation and higher productivity.
- Ultra-high specificity.
- Fast activation.
- Greater efficiency and sensitivity in a wide range of PCR assays within a short reaction time.
- Recommended for low copy assays.
- Amplifies fragments up to 5 kb.
- Unbeatable value in terms of cost per unit and performance.

#### **Applications**

- Hot start PCR.
- Low-copy target.
- High throughput PCR applications
- Complex genomic templates.
- Products suitable for TA cloning

### **Concentration and Components**

GemTaq<sup>TM</sup> Hot Start 5xMaster Mix is a premixed ready-to-use **5x** solution containing GemTaq<sup>TM</sup> Hot Start DNA polymerase, dNTPs, 5x1.5mM MgCl<sub>2</sub> and reaction buffer at optimal concentrations for efficient amplification of DNA templates by PCR.

## **Storage Conditions**

GemTaq<sup>TM</sup> Hot Start 5xMaster Mix can be stored for up to 6 months at -20°C. Repeated freeze/thaw cycles should be minimized.

Note. If precipitation occurs during storage at freezing temperatures, it should be dissolved before usage.

## **Basic PCR Protocol**

# (For amplifications of DNA fragments shorter than 1000 bp)

- 1. Thaw GemTaq<sup>TM</sup> Hot Start 5xMaster Mix completely, mix thoroughly before use and keep it on ice. (If precipitation occurs during storage at freezing temperatures, it should be dissolved before usage.)
- 2. Add nuclease-free water (provided by the user), 0.2-0.4µM of each primer (provided by the user) and template DNA (provided by the user). Mix the content of each PCR sample gently (A).
- 3. If using a thermal cycler without a heated lid, overlay the reaction mix with 1-2 drops of Mineral Oil. Centrifuge the reaction mix in a microcentrifuge for 5 seconds.
- 4. **Guidelines for Thermal Cycling program.** (Researchers can use their own protocols for specific applications.)

1 cycle	Denaturation	94-95 °C	2-3 min
	Annealing and elongation	72 °C	2-4 min
	(Hot Start – user choice),		
20-40 cycles	Denaturation	94-95 °C	15-30 sec
	Annealing	56 °C (B)	15-30 sec
	Elongation	72 °C (C)	0.5-1 min
Post-PCR (to finish	Elongation	72 °C	5 min
extension of all templates)			
Store		4° C	until analysis of samples

### Notes

- (A) Negative controls are recommended for each set of the experiments, when instead of template DNA, an equivalent volume of nuclease-free water is added to one of the samples.
- (B) The annealing temperature for a specific amplification reaction will depend upon the sequences of the two primers and could vary from 50°C to 65°C based on the primer Tm.
- (C) Extension of the primer by GemTaq<sup>TM</sup> Hot Start DNA Polymerase requires approximately 0.5-1 minute per 1000bp of the template to be amplified.

# WARNINGS

We recommend the use of lab coats, gloves and eye protection when working with these reagents. MGQuest assumes no liability for any damage resulting from handling or from contact with the above products.

## LICENSES/PATENTS

A purchase of this product does not include a license to perform any patented application.