#### **INSTRUCTIONS**

### **GM Biosciences**

## Visual-PCR<sup>TM</sup> GFP Tag Detection Kit\* Catalog No. GM7099

1908.05

GM7099 Visual-PCR<sup>™</sup> GFP Tag Detection Kit 15 Tests (1 set of component A, B, C and D)
 Contents: Component A, B and C: to prepare complete-PCR-cocktail
 Component D: synthesized GFP gene for positive control
 Storage: Product shipped on ice, store at -20 °C upon arrival

### Introduction

Visual-PCR<sup>™</sup> GFP Tag Detection Kit is a simple, quick and sensitive way to visually detect GFP gene from plasmids, bacterial colonies, or mammalian cells. It bases on our proprietary Visual-PCR technology. The technology makes PCR amplification to change color. For the very first time, PCR is capable of being visually monitored by naked eyes in visible light. DNA extraction is not required and crude sample is directly used as PCR template. Post-PCR gel electrophoresis is unnecessary.

### Protocol

1. Prepare complete-PCR-cocktail by mixing component A, B and C. Briefly spin tube A, B and C. Mix the whole tube of A (~192ul) and B (~176ul) first, and then add the mixture of A+B into tube C to give a complete-PCR-cocktail. Mix thoroughly and aliquot 24ul of complete-PCR-cocktail for each reaction. A desired complete-PCR-cocktail should be purple or violet, and it contains all the components for PCR reaction. Use the complete-PCR-cocktail within one day. Do NOT freeze it.

#### 2. Add 1ul samples into each reaction.

<u>Bacterial colony</u>: take 1 colony in 100ul water, after vortex, directly take 1ul as template. <u>Plasmid</u>: dilute in water to give a desired concentration<sup>§</sup>, take 1ul as template.

<u>Positive Control and Negative Control</u> are necessary. Add 1ul component D into complete-PCR-cocktail as Positive Control. To rule out the possibility of cross-sample contamination when adding samples, it is suggested to close the tube of Negative Control all the time and add nothing for Negative Control.

- 3. **Perform PCR:** 94 °C 2 min, 50 cycles of (94 °C 5 sec, 70 °C 10 sec). PCR machine equipped with heated lid is necessary.
- 4. Visually check the color of PCR results. After PCR, chill the PCR tube on ice, and then check the color of reaction by naked eyes in visible light. Positive Control and GFP-positive samples are blue or sky-blue. Negative Control and GFP-negative samples<sup>§</sup> are purple or violet.

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Note: Post-PCR gel electrophoresis is not necessary. However, if the sample shows color between purple and blue, agarose gel electrophoresis can be performed to confirm the result. The GFP positive PCR reaction gives a 500bp band.



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§ The detection limit of the kit is 2 copies/reaction.
\* Patent pending

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