

## Product Description

AcuGenix™ HandyAmp Fast PCR Master Mix is a product containing a genetically engineered hot-start DNA polymerase with high amplification success rate. This polymerase possesses extremely high DNA binding affinity and strong processivity, enabling rapid amplification with an extension rate of 1–30 sec/kb ( $\leq 2$  kb: 1 sec/kb,  $\leq 5$  kb: 5 sec/kb,  $\leq 10$  kb: 10 sec/kb). It features broad template compatibility, suitable for genomic DNA or crude templates from animals, plants and bacteria, as well as target sequences with high or low GC content. In addition, this product is pre-formulated as a 2×Premix. Amplification can be performed simply by adding primers and template. A version with pre-loaded electrophoresis loading dye is also available, allowing direct electrophoresis of PCR products for convenient and efficient operation.

## Components

Components	Cat.	Size
2×AcuGenix™ HandyAmp Fast PCR Master Mix <sup>1</sup>	BR1A401-01	5 x 1 mL
	BR1A401-02	25 x 1 mL
	BR1A401-03	50 x 1 mL
2×AcuGenix™ HandyAmp Fast PCR Master Mix (Dye plus) <sup>1,2</sup>	BR1A401-11	5 x 1 mL
	BR1A401-12	25 x 1 mL
	BR1A401-13	50 x 1 mL

1. This reagent already contains hot-start DNA polymerase, PCR Buffer, dNTPs, MgCl<sub>2</sub>, stabilizers, and enhancers, etc.

2. Pre-mixed electrophoresis loading dye is included in this reagent, enabling direct electrophoresis of PCR products.

## Storage

Store at  $-20\pm 5^{\circ}\text{C}$ .

## Notes

- For Research Use Only. Not for use in diagnostic procedures.
- Mix thoroughly before use and avoid repeated freeze-thaw cycles.
- Perform initial denaturation at 95 °C or 98 °C for 3–5 min.
- The fast DNA polymerase enables an amplification rate up to 1 sec/kb.
- To increase PCR yield, appropriately extend the extension time as recommended.
- This product supports 5-plex amplification of fragments up to 10 kb. For multiplex amplification, an extension time of 30 sec/kb is recommended.
- It features excellent system compatibility and is suitable for rapid amplification reactions.
- This product is applicable to fast PCR, genotyping, colony PCR and other experiments.
- When preparing crude animal and plant templates, cut the tissues into small pieces as much as possible to facilitate sufficient DNA lysis.
- If poor amplification is obtained with crude templates, dilute the template appropriately before re-amplification.

## Prepare Reaction Mix

Components	50 $\mu\text{L}$ reaction
2×AcuGenix™ HandyAmp Fast PCR Master Mix	25 $\mu\text{L}$
25× Primer Mix <sup>1</sup>	2 $\mu\text{L}$
Template <sup>2</sup>	--
RNase-free ddH <sub>2</sub> O	To 50 $\mu\text{L}$

1. When using conventional PCR procedure, the final primer concentration of 0.2  $\mu\text{M}$  can get better amplification results, when the reaction performance is poor, the primer concentration can be adjusted in the range of 0.2-1  $\mu\text{M}$ .

2. The copy number of target genes varies among different types of templates. If necessary, perform gradient dilution to determine the optimal template dosage. The addition amount of crude template shall not exceed 1/10 of the total PCR reaction volume.

## Reaction Program

PCR procedures			
Steps	Temp	Time	Cycles
Initial denaturation	95°C	3-5 min	1
Degeneration	95°C	10-15 s	30-35
Annealing <sup>1</sup>	55-65°C	10-30 s	
Elongation	68-72°C	X sec <sup>2</sup>	
Final extension	72°C	3-5 min	1

1. Set the annealing temperature according to the primer T<sub>m</sub> value. If necessary, it is recommended to establish a temperature gradient to identify the optimal temperature for primer-template binding. In addition, annealing temperature directly determines amplification specificity. Non-specific amplification can be reduced by appropriately increasing the annealing temperature.

2. Refer to the extension rate: ≤2 kb at 1 sec/kb, ≤5 kb at 5 sec/kb, ≤10 kb at 10 sec/kb, it is recommended to calculate the extension time based on the longest fragment.

## FAQs

1. Non-specific bands or smearing appear.

- (1) Optimize primer design.
- (2) Increase the annealing temperature or reduce the primer concentration.
- (3) Use purified template.

2. Target band appears in the blank control.

This indicates contamination. Replace the qPCR Master Mix, primers or nuclease-free water and repeat the experiment. Prepare the reaction system in a biosafety cabinet or clean bench to minimize aerosol contamination.

3. Weak or absent target band.

- (1) Optimize primer design.
- (2) Appropriately increase the template input amount.
- (3) Check for primer degradation and ensure the template is intact and undegraded; re-prepare the template and repeat the experiment if needed.
- (4) Verify the correctness of the reaction system and amplification program.