AcuGenix™ Hot Start Colorimetric LAMP Master Mix (Red-to-Yellow)-V2 V01

Product Description

AcuGenix™ Hot Start Colorimetric LAMP Master Mix (Red-to-Yellow)-V2 utilizes visual methods for the qualitative detection of RNA in specimens (e.g., viruses). AcuGenix™ Bst Hot Start DNA Polymerase-V2 is derived from the DNA polymerase of Bacillus stearothermophilus. It exhibits 5′→3′ DNA polymerase activity and strong strand displacement capability, while lacking 3′→5′ exonuclease activity. Compared with the large fragments of wild-type Bst DNA polymerase, this enzyme significantly enhances the amplification speed and yield. It is obtained through reversible modification technology based on genetically engineered Bst 2.0 DNA polymerase. The activity of this enzyme can be completely inactivated at room temperature, allowing reactions to be initiated under ambient conditions. This suppresses non-specific amplification and improves reaction efficiency. This reagent employs an optimized formulation of the LAMP visual assay buffer. It operates on the principle of pH reduction during the amplification reaction, enabling result interpretation through color changes. Before the reaction, the reagent appears purplish-red. After the reaction, positive wells turn yellow, while negative wells remain purplish-red. Additionally, AcuGenix™ Bst Hot Start DNA Polymerase-V2 does not require separate activation steps.

Components

1. AcuGenix™ Bst Hot Start DNA Polymerase-V2 (8 U/μL)
2. 2×LAMP Premix Buffer (RY)

Storage

Store at -20±5℃. Vortex before use. Avoid freeze-thaw cycles.

Prepare Reaction Mix

Components	25 μL System	50 μL System	Final Concentration
AcuGenix™ Bst Hot Start DNA Polymerase-V2 (8 U/μL)	1 μL	2 μL	0.32 U/μL
2×LAMP Lyophilization Buffer (RY)	12.5 μL	25 μL	1×
10×Primer Mix [1]	2.5 μL	5 μL	1×
Template DNA [2]	——	——	——
ddH_2O	To 25 μL	To 50 μL	——

1. Recommended primer concentration: The 10×Primer Mix comprises 16 μM FIP, 16 μM BIP, 2 μM F3, 2 μM B3, 4 μM LoopF, and 4 μM LoopB. Concentration gradient testing can be carried out to determine the optimal primer dosage. It is recommended that primers be prepared with TE Buffer.

2. Different types of DNA templates have different copy numbers of target genes. When necessary, gradient dilution can be performed to determine the optimal quantity of DNA template to be added. This product functions as a pH-sensitive indicator reagent. High-concentration Tris-HCl buffer systems should be avoided in sample elution solutions. The use of strong acid or strong alkali nucleic acid release agents for sample processing should be avoided. If such reagents are unavoidably used, it is advisable to adjust the pH to neutral.

Reaction Conditions

Incubate at a temperature range of 60-65 ℃ for a duration of 30-60 minutes, and acquire fluorescence data at one-minute intervals. After the reaction is completed, allow the sample to cool to ambient temperature before analyzing the results.

Quality control

1. Functional testing: sensitivity, specificity, reproducibility of LAMP.
2. No exogenous nuclease activity, no exogenous endo/exonuclease contamination.

Q & A

LAMP amplification products are scarce or non-existent:

1. Elevate the quantity of the template and the concentration of the enzyme.
2. Substitute the primers. It is advisable to design multiple primer sets and conduct screening for the optimal primers via preliminary experiments. For primer design, refer to http://primerexplorer.jp/e/.
3. The use of Loop Primers is recommended to enhance amplification efficiency and reduce amplification time.

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