

Bst III Fluorescent LAMP Kit (DNA&RNA)

Please read the manual carefully before use.

Cat. No. LP312

Version. No. Version 1.0

Storage: at -20°C for two years.

Description

This product contains *Bst* III Fluorescent LAMP Enzymes Mix, 4×LAMP Reaction Mix I, fluorescent dye TS LAMP Green, and only prepare template and primers. Among them, *Bst* III Fluorescent LAMP Enzymes Mix is an upgraded version of *Bst* II DNA polymerase, which can be used for LAMP reaction with DNA or RNA as template. It is not recommended to add reverse transcriptase when it is used for RT-LAMP reaction. 4×LAMP Reaction Mix I is an optimized reaction mastermix, which already contains MgSO₄, dNTPs and other components required for the reaction, and no additional addition is required.

TS LAMP Green is a DNA-binding dye for fluorescence quantitative amplification. It has a similar spectrum to SYBR Green I and is compatible with all well-known brands of qPCR instruments. Replacing SYBR Green I with TS LAMP Green does not require any changes to current procedures and instrument.

This product is suitable for RT-LAMP reaction with RNA as template, or LAMP reaction with DNA as template, due to strong reverse transcription activity and amplification ability, which can detect RNA or DNA molecules as low as 1 copy within 30 minutes, and can be applied for fluorescent dye method.

Features

- Isothermal Amplification (LAMP/RT-LAMP) capability
- Fast polymerization
- Strong strand-displacement capability

Application

- RNA/DNA isothermal amplification
- DNA sequencing with GC-rich regions
- Applicable for experiments requiring mesophilic strand-displacement

Kit Contents

Component	LP312-01 (100 rxns)	LP312-02 (200 rxns)
<i>Bst</i> III Fluorescent LAMP Enzymes Mix	200 µl	400 µl
4×LAMP Reaction Mix I	0.65 ml	2×0.65 ml
TS LAMP Green (20×)	45 µl	90 µl
6×DNA Loading Buffer	500 µl	1 ml
RNase-free Water	2×1 ml	4×1 ml

Recommended Reaction System based on Fluorescent Dye Method (taking 25 µl reaction system as an example)

Component	Volume	Final Concentration
RNA Template	Variable	≥1 copy
FIP/BIP Primers	Variable	1.6 µM each
F3/B3 Primers	Variable	0.4 µM each
Loop F/B Primers	Variable	0.8 µM each
4×LAMP Reaction Mix I	6.25 µl	1×
TS LAMP Green (20×)	0.45 µl	0.36×
<i>Bst</i> III Fluorescent LAMP Enzymes Mix	2 µl	-
RNase-free Water	Variable	-
Total Volume	25 µl	-



Recommended Reaction Conditions

Incubate at 65°C for 30~45 minutes, and set the read time to 1 minute. The specific reaction temperature is determined according to the T_m value of the primer. It is recommended to incubate at 85°C for 20 minutes after the reaction to inactivate the enzyme.

Operation Suggestions & Notes

- ① Avoid RNase contamination during reaction system preparation;
- ② *Bst* III DNA Polymerase does not lack 5'→3' exonuclease activity, so TaqMan probe method is not recommended;
- ③ *Bst* III DNA polymerase cannot be used for thermal cycle sequencing or PCR;
- ④ Reaction temperature range: 50 °C ~ 65 °C;
- ⑤ Since *Bst* III DNA polymerase is also active at room temperature, please keep it at a low temperature during the preparation of the reaction mix (operate on ice);
- ⑥ The amount of TS LAMP Green dye can be adjusted appropriately, but too high concentration may cause a delay in Ct value;
- ⑦ Please use RNase-free Water or 0.1×TE Buffer to dilute the primers. The buffer concentration in the reaction system is high, and the primers diluted with 1×TE Buffer may affect the amplification;
- ⑧ After preparing the reaction system, it is recommended to add a drop of paraffin oil for liquid seal, which can effectively avoid false positives caused by aerosol contamination, and paraffin oil does not affect the fluorescence value reading results;
- ⑨ Try to separate the experimental environment and prepare the reaction reagents and templates in different areas. If agarose gel electrophoresis or other analysis method that requires opening the LAMP reaction tube is needed after the reaction, please carry out in a separate operating environment to avoid contamination.

For research use only, not for clinical diagnosis

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