

Characteristics

1. This reagent can be used for nucleic acid (both DNA and RNA) extraction, enrichment, and purification from animal samples including whole blood, serum, plasma, urine, saliva, oral swabs, nasopharyngeal swabs, and cell suspensions, or plant samples including roots, stems, leaves, flowers, and seeds. The lysate after nucleic acid release reagent treatment can be directly used for PCR amplification with Tiosbio® Marathon DNA Polymerase (BT0016), and for isothermal amplification with Tiosbio® RAA Nucleic Acid Amplification Kit (JY0200), Tiosbio® RT RAA Nucleic Acid Amplification Kit (JY0203), and Tiosbio® RAA Nucleic Acid Amplification Kit (Lateral Flow Strip Method) (JY0202).
2. No addition of enzymatic preparations such as lysozyme, lysostaphin, or proteinase K is required. No use of harmful chemical reagents such as phenol or chloroform is needed.
3. One-step, single-tube operation that is convenient and fast, with an operation time of less than 1 minute.
4. High sensitivity and good reproducibility. After processing with this kit, taking 1-15 μL of lysate supernatant as template for PCR can stably amplify target genes with an input of 10^1 copies.
5. Can be used for sample pretreatment in food safety, disease control, emerging infectious diseases, animal epidemics, molecular diagnostics, and genetic testing for precision medicine.

Storage Conditions and Shelf Life: 12 months

Specifications and Storage: 1 mL per tube, store at room temperature.

Procedure

1. Animal Samples

After appropriate processing of samples such as whole blood, serum, plasma, urine, saliva, oral swabs, nasopharyngeal swabs, and cell suspensions, mix with the sample to be tested at a ratio of Cool Flash Nucleic Acid Release Reagent (BT0068) to liquid sample = 1:7. When the sample concentration is low, it is recommended that the lysis system be $\geq 50 \mu\text{L}$ to ensure that the lysate contains a detectable amount of nucleic acid sample.

After thoroughly pipetting the mixed solution 20 times with a pipette tip or disposable plastic pipette, aspirate the supernatant into a clean centrifuge tube for testing.

For PCR, RAA, or RT RAA amplification, the lysate supernatant can be added up to the maximum allowable volume of the reaction system.

2. Plant Samples

After simple rinsing of the material to be tested, cut into 2-5 mm square small pieces or segments, add 100 μL of Cool Flash Nucleic Acid Release Reagent (BT0068), and crush the material several times with a pipette tip. After simple crushing of the sample, thoroughly pipette 20 times with a pipette tip or disposable plastic pipette, then aspirate the supernatant into a clean centrifuge tube for testing.

For PCR, RAA, or RT RAA amplification, 1 μL of lysate supernatant can be taken for detection.

Addition amount of lysate in amplification system: If the detection results of the sample to be tested, positive control, and negative control samples after lysis do not match the actual situation, parallel tests can be set up with a gradient of $\pm 2 \mu\text{L}$ of lysate addition volume to determine the effect of different lysate volumes on the detection of samples, positive controls, and negative controls within the same amplification system. Under the condition that the positive control detection result is positive and the negative control detection result is negative, select the maximum lysate addition amount. Since Cool Flash Nucleic Acid Release Reagent has the effect of inhibiting non-specific amplification, when all negative controls are detected as negative and all positive controls are detected as positive, the lysate can be added at the maximum sample addition volume allowed by the amplification system.

Warnings and Precautions

1. When using this reagent for nucleic acid extraction from whole blood, serum, plasma, urine, saliva, oral swabs, nasopharyngeal swabs, or cell suspensions, sample processing should be completed immediately after sample collection whenever possible. If conditions do not allow, samples should be transported under refrigerated or frozen conditions.

2. When used for infectious disease sample testing, pay attention to safety protection during operation. Operations should be conducted in designated laboratory facilities. Wear protective clothing, disposable gloves, and masks. All items that have directly contacted pathogen samples should be disinfected before disposal or reuse.

3. This reagent is designed for rapid crude processing of animal liquid samples. The processed material can be

used for PCR amplification with Tiosbio® Marathon DNA Polymerase (BT0016), and for isothermal amplification with Tiosbio® RAA Nucleic Acid Amplification Kit (JY0200), Tiosbio® RT RAA Nucleic Acid Amplification Kit (JY0203), and Tiosbio® RAA Nucleic Acid Amplification Kit (Lateral Flow Strip Method) (JY0202). The processed material is not suitable for experiments requiring high nucleic acid purity, such as UV spectrophotometric quantification.

4. When lysing samples with Cool Flash Nucleic Acid Release Reagent (BT0068), the Cool Flash Nucleic Acid Release Reagent (BT0068) is an 8 × solution, and the lysis system is Cool Flash Nucleic Acid Release Reagent (BT0068) to liquid sample = 1:7. This ratio has been repeatedly optimized. Do not adjust arbitrarily!

5. For PCR, RAA, or RT RAA amplification, the processed supernatant can be added up to the maximum allowable volume of the reaction system. When the amplification template is DNA, the stored processed supernatant at -20°C can be reused multiple times within 24 hours. The success of PCR or RAA reactions is closely related to primer design, cycling parameters, and amplification time settings. If amplification fails after processing samples with this kit, please confirm whether PCR or RAA reactions were successful using nucleic acid extracted by other methods.

6. This reagent is for research use only. Do not use for clinical, food, or other purposes.