Universal Plant Total RNA Kit

Catalog No.: abx298034

Size: 50 reactions / 200 reactions

Storage: Store the DNAse I enzyme at -20°C. Store all other components at room temperature in a dry environment.

Application: For isolation of total RNA content from plant tissue samples.

Introduction

Abbexa's Universal Plant Total RNA Kit is a quick and convenient method for isolating total RNA content from fresh and dried plant tissue samples, including samples rich in polysaccharides and polyphenols. The membrane technology used in the kit allows the removal of secondary metabolites such as polysaccharides, polyphenols, and lipids without the need for hazardous reagents such as phenol or choloroform. Purified RNA recovered with this kit is suitable for RT-PCR, qRT-PCR, Northern Blot, and other applications.

Kit components

1. Lysis Buffer: 40 ml / 160 ml
2. Precipitation Buffer: 10 ml / 40 ml
3. DNAse I (3 U/µl): 1500 U / 6000 U
4. DNAse I Reaction Buffer: 4 × 1 ml / 15 ml
5. Clean Buffer: 20 ml / 80 ml
6. Wash Buffer: 12 ml / 2 × 22 ml
7. RNAse-free Water: 10 ml / 20 ml
8. Filtration Column with Collection tube : 50 / 200
9. RNA Spin Column with Collection tube: 50 / 200

Materials required but not provided

1. Isopropanol
2. Absolute Ethanol
3. Pipette and sterile pipette tips
4. Sterile 1.5 ml microcentrifuge tubes
5. Centrifuge
6. Incubator

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Protocol

A. Preparation of reagents

- **DNAse I Working Solution:** Add 70 µl DNAse I Reaction Buffer to an RNAse-free microcentrifuge tube, then add 10 µl DNAse I enzyme and mix thoroughly.
- **Clean Buffer:** Add 20 ml **(50 rxns)** or 80 ml **(200 rxns)** absolute ethanol before use.
- **Wash Buffer:** Add 48 ml **(50 rxns)** or 2 × 88 ml **(200 rxns)** absolute ethanol before use.

Notes:

- Samples should be fresh with minimal freeze-thaw cycles.
- Increase initial sample mass for high water content samples such as fruit tissue.
- Always use sterile pipette tips and tubes to avoid RNAse contamination.
- If Lysis Buffer has precipitate, incubate in a 37 °C water bath until clear.

B. Extraction Procedure

1. Weigh approximately 100 mg of fresh plant tissue, or 30 mg of dried tissue into a sterile microcentrifuge tube.
   Note: Dried samples should be ground with liquid Nitrogen.
2. Add 700 µl Lysis Buffer and mix thoroughly.
3. Incubate at 65°C for 5 minutes.
4. Add 175 µl Precipitation Buffer and mix thoroughly.
5. Centrifuge at 13,500 × g for 3 minutes, then transfer the supernatant to a Filtration Column with collection tube and centrifuge at 13,500 × g for 2 minutes.
   Note: If solution is viscous after lysis, place in an ice bath for 5 minutes before the first centrifuge step.
6. Add 350 µl Isopropanol and pipette up and down to mix. Flocculent precipitates may appear in this phase.
7. Transfer solution to an RNA Spin Column with collection tube, then centrifuge at 13,500 × g for 30 seconds. Discard the flowthrough.
8. Add 80 µl DNAse I Working Solution to the center of the filter membrane in the spin column and stand at room temperature for 10 minutes.
9. Add 500 µl prepared Clean Buffer and centrifuge at 13,500 × g for 30 seconds. Discard the flowthrough.
10. Add 500 µl prepared Wash Buffer and centrifuge at 13,500 × g for 30 seconds. Discard the flowthrough.
11. Repeat Wash Buffer step detailed in step 10.
12. Centrifuge at 13,500 × g for 2 minutes to remove residual Wash Buffer, then transfer the spin column to a fresh microcentrifuge tube.
13. Add 50-100 µl RNAse-free water to the center of the spin column, then stand at room temperature for 2 minutes.
14. Centrifuge at 13,500 × g to elute the RNA into the microcentrifuge tube.
   Note: Step 13 and 14 may be repeated once for additional RNA yield.

C. Technical Support

For troubleshooting and technical assistance, please contact us at support@abbexa.com.

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