

EasyPure[®] Plant RNA Kit

Cat. No. ER301

Storage: DNase I and DNase I Reaction Buffer at -20°C for one year; others at room temperature (15-25°C) for one year

Description

EasyPure[®] Plant RNA Kit provides a simple and fast column based method to isolate RNA from plant tissue. Samples are lysed with detergent to inactivate RNase. DNA is digested with DNase I. RNA is bound to silica membrane. After washing, high quality RNA is eluted from the column. RNA is free of protein contamination, and is suitable for RTPCR, qRT-PCR, Microarray analysis and Northern blot.

Kit Contents

Component	ER301-01 (50 rxns)
Binding Buffer 6 (BB6)	60 ml
Wash Buffer 6 (WB6)	12 ml
Clean Buffer 6 (CB6)	60 ml
DNase I (3 units/μl)	1500 units
DNase I Reaction Buffer	4×1 ml
RNase-free Water	10 ml
RNase-free Tube (1.5ml)	50 each
RNA Spin Columns with Collection Tubes	50 each

Sample Requirement

Material	Volume of BB6/β-ME
≤ 100 mg	0.5 ml
100-200 mg	1 ml

Procedures

Before starting, add 48 ml 96-100% ethanol to WB6, mix thoroughly.

1. Materials preparation

Grind proper amount of plant tissues (refer to sample requirement) quickly in liquid nitrogen, add volume of BB6 containing β-mercaptoethanol (10 μl of β-mercaptoethanol/ml BB6, freshly prepared), mix thoroughly by vortexing. Incubate for 3 minutes at room temperature.

2. Centrifuge at 12,000×g for 2-5 minutes and gently transfer the supernatant to a clean RNase-free microcentrifuge tube.

3. Add half volume of 96-100% ethanol to the supernatant and mix well (visible precipitates may form at this moment).

4. Vortex to disperse the precipitates.

5. Transfer the sample (including any remaining precipitate) to a spin column. Centrifuge at 12,000×g for 30 seconds. Discard the flow-through (repeat this step if the volume of the sample is larger than the volume of the spin column can hold).

6. Add 500 μl of CB6 to the spin column. Centrifuge at 12,000×g for 30 seconds. Discard the flow-through.

Optional: When genomic DNA-free RNA is needed, add 80 μl of DNase I working solution (the working solution is prepared by mixing 70 μl of reaction buffer and 30 U DNase I) and incubate for 15 minutes at room temperature.

7. Repeat step 6 once without DNase digestion.

8. Add 500 μ l of WB6 (check to make sure that ethanol has been added) to the spin column. Centrifuge at 12,000 \times g for 30 seconds. Discard the flow-through.
9. Repeat step 8 once.
10. Centrifuge at 12,000 \times g for 2 minutes at room temperature to remove ethanol and residue. Air-dry the column matrix for several minutes.
11. Place the spin column into a clean 1.5 ml RNase-free tube. Add 30-100 μ l of RNase-free Water into the center of spin column and incubate at room temperature for 1 minute.
12. Centrifuge at 12,000 \times g for 2 minutes to elute RNA.
13. Store the isolated RNA at -80°C.

Notes

- All the centrifugation steps are carried out at room temperature.
- Ensure to add β -mercaptoethanol to BB6 before use.
- Ensure to add 96-100% ethanol to WB6 before use.
- The kit is suitable for RNA purification from normal plant tissues, not suitable for RNA purification from plant tissues rich in polysaccharide or polyphenol.
- We recommend to use *TransZol Plant* for RNA purification from plant tissues rich in polysaccharide and/or polyphenol.

FOR RESEARCH USE ONLY