Version: 12/2023

5. Add 350 μL of Buffer RW1 to the Spin Columns RM, centrifuge at 12,000 rpm for 1 min at 4°C, discard the waste solution, and return the column to the collection tube.
6. Prepare DNase I mixture: Take 52 μL of RNase-Free Water, add 8 μL of 10× Reaction Buffer and 20 μL of DNaseI (1 U/μL) to it, mix well, and prepare a final volume of 80 μL of reaction solution.
7. Add 80 μL DNase I mixture directly to the adsorption column and incubate at 20-30°C for 15 min.
8. Add 350 μL Buffer RW1 to the Spin Columns RM and centrifuge at 12,000 rpm for 1 min at 4°C. Discard the waste solution and put the adsorbent column back into the collection tube.
9. Add 500 μL Buffer RW2 (please check whether 100% ethanol is added before use) to the Spin Columns RM, centrifuge at 4°C 12,000 rpm for 1 min, discard the waste solution and return the column to the collection tube.
10. Repeat step 9.
11. Centrifuge at 4°C 12,000 rpm for 2 min.

    Note: The purpose of this step is to remove residual ethanol from the adsorbent column; ethanol residue can interfere with subsequent enzymatic reactions (zymography, PCR, etc.).
12. Load the Spin Columns RM into new RNase-Free Centrifuge Tubes (1.5 mL), add 30-50 μL of RNase-Free Water dropwise to the middle of the adsorbent membrane overhanging the adsorbent membrane, leave it at room temperature for 2 min, and centrifuge it at 4°C 12,000 rpm for 1 min, and store the obtained RNA solution at -70°C to prevent degradation.

    Note: 1) The volume of RNase-Free Water should not be less than 30 μL; too small a volume affects the recovery rate.

    2)If you want to increase the yield of RNA, repeat step 12 with 30-50 μL of new RNase-Free Water.

    3)If you want to increase the RNA concentration, the obtained solution can be re-added to the adsorption column and repeat step 12.

OminiPlant RNA Kit （Dnase I）

Cat. No. : CW2598S (50 preps)

Storage Condition: DNase I and 10× Reaction Buffer are stored at-20°C, and other components are stored at room temperature (15-30°C).

Components

Component	CW2598S (50 preps)
DNase I	1000 U
10× Reaction Buffer	1000 μL
Buffer RLS	40 mL
Buffer RW1	40 mL
Buffer RW2 (concentrate)	11 mL
RNase-Free Water	10 mL
Spin Columns FS with Collection Tubes	50
Spin Columns RM with Collection Tubes	50
RNase-Free Centrifuge Tubes（1.5 mL）	50

Introduction

This kit is suitable for extracting RNA from a wide range of plants, even from plants rich in polysaccharides and polyphenols, high quality RNA can be successfully extracted, such as rice leaves, wheat leaves, corn leaves, tobacco leaves, pine needles, ginkgo leaves, poplar leaves, pomegranate leaves, holly leaves, apples, peaches, pears, tomatoes, cherries, apricots, bananas, grapes, loquats, cinnamon rinds, cinnamon pulp, lychee fruit rinds, lychee pulp, soybean, peanut, corn, potato tuber, moonflower petal, pomegranate petal, shiitake mushroom, flat mushroom and other samples. The unique lysate formulation can rapidly inactivate the RNA enzyme in the cell, effectively remove the effect of polysaccharide and polyphenol on RNA extraction without the need of phenol, chloroform and other reagents, and at the same time, it adopts the silica matrix membrane adsorption of RNA for purification, and the extracted total RNA has high purity and is free of contamination of genomes, proteins and other impurities, and it can be used for Real Time RT-PCR and RT-PCR, Northern Blot, Dot Blot, and in vitro translation and other downstream experiments.

RNA yield

Plant samples	Total RNA yield (μg)
Arabidopsis pods	~50
Soybean	~55
Corn leaves	~55

Reagents to be Supplied by user

β-mercaptoethanol, 100% ethanol (freshly opened or for RNA extraction)

Precautions

1. To prevent RNase contamination, the following aspects should be noted:
   1) Use RNase-free plastic products and pipette tips to avoid cross-contamination.
   2) Operators should wear disposable masks and gloves, and gloves should be changed frequently during the experiment.
2. Samples should avoid repeated freezing and thawing, otherwise the yield and quality of RNA extraction will be affected.
3. If Buffe RLS shows precipitation, heat to dissolve it and leave it at room temperature.
4. Add β-mercaptoethanol to Buffer RLS before use, 1 mL Buffer RLS with 20 μL β-mercaptoethanol. Buffer RLS with β-mercaptoethanol can be stored for 1 month at room temperature.
5. Before using Buffer RW2 for the first time, 100% ethanol should be added according to the instructions on the label of the reagent bottle.

Protocol

1. Homogenate treatment: Take 50-100 mg of plant tissue and quickly grind it into powder in liquid nitrogen, add 500 μL Buffer RLS (please check whether β-mercaptoethanol is added before use), and immediately vortex with vigorous shaking to mix.
   Note: For materials with extremely rich water content, such as watermelon pulp, tomato, pear pulp, etc., more material can be added appropriately, up to 200 mg; for starch-rich samples or mature leaves, the amount of Buffer RLS can be increased appropriately, up to 700 μL.
2. Centrifuge at 12,000 rpm (~13,400 ×g) at 4°C for 2 min.
3. Transfer the supernatant to a Spin Columns FS with collection tube, centrifuge at 12,000 rpm for 1 min at 4°C. Carefully aspirate the supernatant from the collection tube and transfer it to a new RNase-Free centrifuge tube (self-provided), avoiding the tip of the pipette from touching the cell debris precipitate in the collection tube.
4. Slowly add 0.5 times the volume of the supernatant in 100% ethanol, mix well (precipitate may appear at this time), and transfer the resulting solution and precipitate together into a Spin Columns RM with collection tube, or if you can not add all of the solution to the adsorbent column at one time, please transfer it into the adsorbent column in two times. Centrifuge at 12,000 rpm for 1 min at 4°C, discard the waste solution and put the adsorbent column back into the collection tube.