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FASTeasy Universal Plant & Fungi RNA Kit

Product Number: RNK6901

Shipping and Storage

1. Inappropriate storage at low temperatures (4℃ or -20℃) may cause solution precipitation, affecting the effectiveness of use. Therefore, transportation and storage are carried out at room temperature (15℃-25℃).
2. To avoid the volatilization, oxidation, and pH changes of reagents exposed to air for a long time, the lid of each solution should be promptly closed after use.

Components

Component	50 Preps
Buffer FEA	30mL
PlantAid	3mL
Buffer RW1	40mL
Buffer RW	10mL
	Add anhydrous ethanol according to the label instructions before the first use
RNase-free H_2O	5mL
DNA clearance/RNA adsorption universal column and collection tube	100

Note: This reagent kit can be stored at room temperature for 12 months without affecting its effectiveness.

Description

This product is suitable for rapid extraction of RNA from common plants, common polysaccharide polyphenol plants, and fungi. The exclusive development of genomic DNA clearance/RNA adsorption universal column technology combined with special reagent formula generally does not require DNA enzyme digestion, effectively removing gDNA residue, and obtaining RNA without significant DNA residue. It can be directly used for downstream reverse transcription fluorescence quantitative PCR or high-throughput sequencing library construction experiments.

Application

Suitable for rapid extraction of RNA from common plants, common polysaccharide and polyphenol plants, fungi, and other sources.

Features

1. Completely avoid using toxic reagents such as phenol, chloroform, and beta mercaptoethanol, and do not require steps such as ethanol precipitation.
2. Simple and straightforward, a single sample operation can generally be completed within 12 minutes.
3. Exclusive research and development of genomic DNA clearance/RNA adsorption universal column technology can effectively remove gDNA residues, resulting in extremely high RNA purity, with a typical OD260/OD280 ratio of 2.1~2.2. Generally, DNase digestion is not required and can be used for experiments such as reverse transcription PCR, fluorescence quantitative PCR, high-throughput sequencing library construction, etc.
4. The world's leading adaptability is extremely extensive, successfully extracting hundreds of samples including cotton, roses, Arabidopsis, rice, tobacco, poplar, etc., including some samples that failed to be extracted by domestic and foreign reagent kits. If the effect of particularly complex polysaccharide polyphenols and other samples is not satisfactory, or if other companies fail to produce particularly difficult samples, EASYspin Plus Complex Polysaccharide Polyphenols Plant RNA Extraction Kit can

be selected.

Note

1. All centrifugation steps can be completed at room temperature. A traditional desktop centrifuge with a speed of up to 13000 rpm can be used.
2. You need to bring your own ethanol and mortar (optional).
3. Buffer FEA and Buffer RW1 contain irritating compounds. Wear latex gloves during operation to avoid contact with skin, eyes, and clothing. If it comes into contact with the skin or eyes, rinse with plenty of water or saline solution.
4. This kit can remove most of the DNA residues in the system, and the purified RNA can usually be used for downstream experimental operations without the need for DNase I treatment. The nucleic acid content of different samples varies greatly. If downstream experiments are highly sensitive to trace amounts of DNA, DNase I can be used to further remove DNA contamination or a DNase I column digestion step can be added during the extraction process.

Protocol

Before the first use, please add anhydrous ethanol according to the instructions on the label of the Buffer RW bottle!

1. Take 550μL Buffer FEA into a 1.5mL centrifuge tube and add 55μL PlantAid for later use.
2. After grinding an appropriate amount of plant/fungal tissue into fine powder in liquid nitrogen, take 50 mg-100 mg of fine powder and transfer it into the centrifuge tube containing Buffer FEA (already added PlantAid). Immediately vortex vigorously for 30 seconds to fully mix the sample with the lysis solution and complete the lysis. Centrifuge at 13000 rpm for 5-10 minutes.
   The sample processing capacity can be increased or decreased according to specific circumstances, for example, if there is a lot of moisture in the sample, the processing capacity can be appropriately increased.
   PlantAid helps remove polysaccharide polyphenol impurities, which can be omitted from samples of common plants such as rice leaves. In some cases, if the RNA production concentration is low, it is also possible to try not adding PlantAid. In some cases, not adding PlantAid may increase the RNA production concentration and success rate.
3. Immediately take about 500μL of supernatant and transfer it to a DNA clearance/RNA adsorption universal column (already placed in a collection tube, hereinafter referred to as the universal column). Centrifuge at 13000 rpm for 30 seconds, discard the universal column, and retain the filtrate in the collection tube (RNA in the filtrate).
   The volume of the supernatant can be adjusted according to the actual situation.
4. Add 0.5 times the volume of the filtrate in anhydrous ethanol (approximately 250μL, adjusted according to the actual situation of the supernatant) to the collection tube, and mix well by pipetting.
   If turbidity or flocculent substances appear after adding alcohol, it is a normal phenomenon. The mixed solution (including flocculent substances) can be added to the universal column for further operation.
5. Immediately transfer the above mixture to a new universal column (already placed in the collection tube), let it stand for 1 minute, centrifuge at 13000 rpm for 30 seconds, discard the filtrate, and place the universal column in the recovery header.
   The adsorption column has a volume of 750μL. If the mixed solution exceeds this volume, please load the column multiple times.
6. Add 700μL of Buffer RW1 to the universal column, let it stand at room temperature for 1 minute, centrifuge at 13000 rpm for 30 seconds, and discard the filtrate.
7. Add 500μL of Buffer RW to the universal column (please check if anhydrous ethanol has been added before use), centrifuge at 13000 rpm for 15 seconds, and discard the filtrate.
8. Repeat step 7 once.
9. Place the universal column in the recycling manifold and centrifuge at 13000 rpm for 2 minutes.
   The air shaking effect is to remove the rinsing solution as much as possible to prevent residual ethanol in the rinsing solution from inhibiting downstream reactions.
10. Transfer the universal column to a new 1.5mL centrifuge tube, suspend 30-50μL of RNase free ddH_2O in the center of the adsorption column membrane, let it stand for 1 minute, and centrifuge at 13000 rpm for 1 minute.

The recommended elution volume is not less than 30μL, as a small volume can affect the efficiency of nucleic acid recovery.

The following steps can help increase the concentration of RNA products: RNase free ddH_2O is preheated at 90℃; Re add the first eluent to the adsorption column for elution, and two washings can increase the concentration by about 10%.

11. The extracted RNA can be directly used for downstream experiments or stored at -85℃ to -65℃.

Appendix : Quick extraction operation steps

Note: For species with low DNA content such as cotton, tomato, and rapeseed, step 3 of the filtration process can be omitted and extracted using the following rapid extraction steps.

Before the first use, please add anhydrous ethanol according to the instructions on the label of the Buffer RW bottle!

1. Take 550μL Buffer FEA into a 1.5mL centrifuge tube and add 55μL PlantAid for later use.
2. After grinding an appropriate amount of plant/fungal tissue into fine powder in liquid nitrogen, take 50mg-100mg of fine powder and transfer it into the centrifuge tube containing Buffer FEA (already added PlantAid). Immediately vortex vigorously for 30 seconds to fully mix the sample with the lysis solution and complete the lysis. Centrifuge at 13000 rpm for 5-10 minutes. Transfer approximately 500μL of supernatant to a new centrifuge tube.

   Take 20μL of DNase I buffer and 2μL of RNase free DNase I centrifuge tube and gently blow and mix them together to form the working solution (when processing multiple centrifuge columns, the working solution should be prepared by scaling up according to the proportion).
3. Add 0.5 times the volume of the filtrate in anhydrous ethanol (about 250μL, adjusted according to the actual situation of the supernatant) to the centrifuge tube, and mix well by pipetting.

   If turbidity or flocculent substances appear after adding alcohol, it is a normal phenomenon. The mixed solution (including flocculent substances) can be added to the universal column for further operation.
4. Follow steps 5-11 to complete the subsequent steps.