ARPENspin Plant Genomic DNA Extraction Kit

Cat# ARP-PGDES50

Kit Components

COMPONENTS	
LP Buffer	22.5mL
LP plus Buffer	22.5mL
DA Buffer	7.5mL
P Binding Buffer	14mL
G Binding Buffer	25mL
Wash Buffer	25.2mL
Elution Buffer	10mL
Spin Columns	50

Storage

• The kit should be stored at 15 - 25°C.

• All reagents, when stored properly, remain stable for up to 18 months.

Introduction

The kit offers a simple, fast, and cost-effective method for isolating pure high-molecular-weight genomic DNA from plant tissues, utilizing the Genomic DNA Buffer Set. This purification procedure takes advantage of the selectivity of the Biospin membrane, enabling the extraction of high yields of pure genomic DNA in less than an hour. It does not require expensive equipment, involves only a few steps, and completely eliminates the need for toxic and hazardous reagents like phenol and chloroform. Typically, 1-30 µg of genomic DNA can be obtained from up to 100 mg of tissue using this kit.

The purified DNA can be extensively utilized in PCR, real-time PCR, sequencing, Southern blotting, mutant analysis, SNP analysis, and other applications.

Additional apparatus and materials required but not supplied

- Sterile 1.5mL microcentrifuge tubes
- 10μL/200μL/1000μL tips
- Centrifuge capable of 14,000g
- Absolute ethanol
- Vortex mixer
- 100mg/ml RNase A

Important Notes

1. Please add 37.8 mL of absolute ethanol to the Wash Buffer and mix thoroughly before first use.
2. Please add 28 ml of absolute alcohol to the P Binding Buffer and mix thoroughly before its first use.
3. LP Buffer can form precipitates during storage. If precipitates form, incubate the buffer at 37°C until completely dissolved.

Protocol

1. Grind the plant tissue into a fine powder while using either liquid nitrogen or an ice bath.
2. Transfer up to 100mg tissue to a 1.5 or 2.0ml microcentrifuge tube. Note: the grinded degree of sample will be affect cell lysis.
3. Add 450 μL of LP Buffer. Optionally, you can add 4 μL of a 100 mg/mL solution of RNase A. Mix thoroughly. If the plant tissues are high in polysaccharides and polyphenols, please use the LP Plus buffer.
4. Incubate at 65°C for 15 minutes, vortexing the tube 2-3 times during incubation. If the sample is difficult to lyse, consider extending the incubation time.
5. Add 150 μL of DA Buffer and mix thoroughly. Then, incubate the mixture on ice for 5 minutes..

6. Centrifuge the lysate at 12,000 × g for 5 minutes, and transfer the supernatant to a new 1.5 ml tube.
7. Add 750 µL (equivalent to 1.5 volumes) of the P Binding Buffer. Mix thoroughly.
8. Transfer the mixture to the spin column and centrifuge at 10,000 × g for 1 minute. Discard the flow-through. If the sample volume exceeds 750 µl, simply load and centrifuge again..
9. Add 500 µL of the G Binding Buffer to the spin column. Centrifuge at 10,000 × g for 30 seconds, then discard the flow-through.
10. Add 600 µL of Washing Buffer to the spin column. Centrifuge at 10,000 × g for 30 seconds, then discard the flow-through.
11. Please repeat step 10th.
12. Centrifuge for an additional minute at 10,000 × g, then transfer the spin column to a sterile 1.5 ml microcentrifuge tube.
13. Add between 100 µL and 200 µL of Elution Buffer. Incubate at room temperature for 1 minute.
14. Centrifuge the microcentrifuge tube at 12,000 × g for 1 minute. The buffer in the tube contains the DNA.
15. The purified DNA can be directly used for various downstream molecular biology experiments. If not used immediately, store it at -20℃.

FAQ

Q1: From which type of plant tissue can we extract DNA using the kit?

A: We can extract DNA from leaves, flowers, caudexes (the bark parts that contain cells), roots (known as pilorhiza), fruits, and seeds (like soybeans and corn).

Q2: When should the sample be centrifuged before filtration?

A: Some samples will become very dense when placed in the DA Buffer ice bath. We can transfer the clear liquid above into the shredder Spin Column after centrifugation.

Q3: What steps can we take when the yield of DNA extraction is low?

A: DNA yield is closely related to lysis efficiency. We can enhance lysis efficiency by extending the incubation time (we can carry out overnight lysis incubation for some samples), which in turn improves DNA yield.

Q4: How can I remove the RNA from the extracted DNA?

A: We can add RNase A to digest the RNA, following the method listed in the specification.

Q5: How long are the extracted genomic DNA fragments?

A: The length of the extracted genomic DNA fragments typically ranges from about 30 to 50 kilobases (KB).