

Product components

Components	Component number	Size		Storage
		50	RXN	
RNase A (10 mg/mL)	RM30149	550	µL	RT
Buffer DL1 Plus	RM30150	25	mL	RT
Buffer DL2	RM30151	10	mL	RT
Buffer DL4	RM30152	21	mL	RT
Buffer EB2	RM30153	15	mL	RT
Buffer DW2	RM30154	12	mL	RT
Spin Column 4	RM30187	50	pk	RT
2 mL Collection Tubes	RM30188	50	pk	RT

Product Description

This kit is suitable for rapid and simple extraction of genomic DNA from various fresh plant tissues or cultured plant cells. The extracted genomic DNA fragments are large with high purity and stability and can be directly used for PCR, digestion, hybridization and others. The extraction process does not require phenol/chloroform extraction. Fresh or frozen plant tissue cells are ground and then lysed and centrifuged by lysate. Proteins, polysaccharides and cell fragments are precipitated and removed by centrifuge at high speed. Under the optimized binding conditions, genomic DNA can be specifically bound to silica membrane of Spin Columns 4. Then, washing steps can further remove the residual proteins, salts and other impurities, and finally DNA is eluted from the silica membrane into Buffer EB2.

Storage Conditions

The kit can be stored at room temperature (15-25 °C) for up to 12 months. Check whether there is any crystallization or precipitation in Buffer DL1 Plus and Buffer DL2 before use. If any, please re-dissolve Buffer DL1 and Buffer DL2 in a 37°C water bath.

Product Features

1. There is no need to use toxic reagents such as phenol/chloroform, mercaptoethanol or DTT, and the extraction process is safe and non-toxic.
2. Simple and convenient operation, no need to water or ice baths, single tube extraction can be completed within 30 minutes at room temperature.
3. Widely applicable and the extraction of various plant samples has been verified.
4. Obtaining high-quality and pure nucleic acid that meets various downstream experimental needs.

Precautions

1. The sample should avoid repeated freeze-thaw, otherwise it may result in smaller extracted DNA fragments and a decrease in extraction volume.
2. Buffer DL1 plus may turn yellow and does not affect the extraction effect.
3. If there is precipitation in Buffer DL1 plus or Buffer DL2, it can be dissolved in a water bath at 37°C, it can be used after shaken well.
4. All centrifugation steps are completed at room temperature in a desktop centrifuge.
5. For polysaccharides and polyphenolics-rich plants, they are recommended to add 2% PVP-40 and 0.2% β- Mercaptoethanol (self prepared) in Buffer DL1 plus.

Operating Procedures

Self prepared materials

Absolute ethanol, 1.5 mL sterile centrifuge tubes (optional: PVP-40, β - Mercaptoethanol).

Preparation before experiment

Before first use, please add 27 mL of absolute ethanol to Buffer DL4 bottle and 48 mL of absolute ethanol to Buffer DW2 bottle.

Operating steps (please read the precautions first)

1. Sample processing: Take fresh or dry plant tissue, add liquid nitrogen, and thoroughly grind it into powder.

Note:

- a. When extracting DNA, the dosage of each tube of sample: fresh plant tissue \leq 100 mg; Dry plant tissue \leq 20 mg. Please select the appropriate amount of plant tissue for liquid nitrogen grinding based on the downstream required amount of DNA.
 - b. If the sample after liquid nitrogen grinding is not immediately used, please store it at -70°C to avoid repeated freeze-thaw.
2. Transfer 100 mg of freshly ground plant tissue or 20 mg of dried plant tissue to a 1.5 mL centrifuge tube (self prepared), and immediately add 400 μL Buffer DL1 plus and 10 μL RNase A (10 mg/mL), vortex oscillate for 2 minutes until the sample and solution are fully mixed, and leave at room temperature for 10-15 minutes.

Note:

- a. The sample and solution must be thoroughly mixed to prevent the sample from clumping in the solution and affecting the final yield of genomic DNA extraction.
 - b. The sample processing capacity should not exceed the capacity of the reagent kit, otherwise it may lead to insufficient sample lysis. For samples with high moisture content, such as strawberries, watermelons, etc., the sample size can be increased appropriately.
 - c. When the content of polysaccharides and polyphenols in the sample is particularly high, 2% PVP-40 and 0.2% β -Mercaptoethanol can be added simultaneously to Buffer DL1 plus .
3. Add 130 μL Buffer DL2, vortex oscillate for 1 minute, thoroughly mixed. Centrifuge at 12000 rpm (13400 \times g) for 5 minutes, carefully suck an appropriate amount of supernatant into a new centrifuge tube.
 4. Add 1.5 times the volume of the supernatant to Buffer DL4 (add 27 mL of absolute ethanol to the bottle for the first use), for example: add 600 μL Buffer DL4 to 400 μL supernatant), immediately shake thoroughly and mix for 30 seconds, at which point flocculent precipitation may occur.
 5. Transfer the obtained solution and precipitate together into the Spin Column 4 (placed in the collection tube). Centrifuge at 12000 rpm (13400 \times g) for 30 seconds, discard the filtrate, and place the adsorption column 4 in the recovery header. (The volume of the adsorption column is about 700 μL , the solution can be added in batches and centrifuged.
 6. Add 500 μL Buffer DW2 (48 mL of absolute ethanol added to the bottle for the first use) to the Spin Column 4, Centrifuge at 12000 rpm (13400 \times g) for 30 seconds, discard the waste liquid, and place the adsorption column in the recovery header.
 7. Repeat step 6 once.
 8. Centrifuge at 12000 rpm (13400 \times g) for 3 minutes and discard the waste liquid from the collection tube.
 9. Transfer the Spin Column 4 into a new 1.5 mL centrifuge tube (self prepared), open the cover of the Spin Column 4, and add 50-200 μL Buffer EB2 suspended droplets to the center of the adsorption membrane, centrifuge at 12000 rpm (13400 \times g) for 1 minute to obtain a DNA solution.

Note:

To increase the yield of genomic DNA, the solution obtained by centrifugation can be added to the adsorption column Spin Column 4, placed at room temperature for 2 minutes, and centrifuged at 12000 rpm (13400 \times g) for 1 minute. Buffer EB2 volume should not be less than 50 μL .