

Genomic DNA Isolation Kit (Plant)

Cat No. PDC10-0100(S)

Size: 100 Reactions

Sample: up to 100 mg of fresh plant tissue
up to 50 mg of dry plant tissue

Format: spin column

Column capacity: up to 50 µg

Operation time: within 60 minutes



Description

The **Genomic DNA Isolation Kit (Plant)** is designed specifically for genomic DNA isolation from plant samples. This unique buffer system ensures total DNA with high yield and good quality from samples. The spin column system is designed to purify or concentrate DNA products which have been previously isolated using buffers. The entire procedure can be completed in 1 hour without phenol / chloroform extraction. Purified DNA is suitable for use in PCR or other enzymatic reactions.

Feature

- Delivering high-quality genomic DNA with the fast procedure
- Ready-to-use genomic DNA for high performance in any downstream application
- Highly purified and high yield genomic DNA can be extracted from various tissue samples.
- Optimized tissue lysis buffer for the efficient lysis.
- Designed to rapidly purify high-quality DNA using spin column format

Kit Contents

Contents	PDC10-0100	PDC10-0100S
Buffer PL	55 ml	2 ml
Buffer W1	45 ml	2 ml
Buffer W2 (Add ethanol)	15 ml (60 ml)	300 µl x2 (1.2 ml x2)
Buffer RE	10 ml	1 ml
Column PC	100 pcs	4 pcs
Collection Tubes	100 pcs	4 pcs

Application

- Gene cloning
- Southern blotting
- PCR
- SNP genotyping

Quality Control

The quality of the Genomic DNA Isolation Kit (Plant) is tested on a lot-to-lot basis to ensure consistent product quality.

Required Materials

- RNase A (50 mg/ml)
- Absolute ethanol
- Isopropanol
- Mortar and pestle
- Microcentrifuge tubes

Genomic DNA Isolation Kit (Plant) Protocol

Step 1 Sample Preparation

1. Cut off 50 mg of the fresh plant tissue or 25 mg of the dry plant tissue.
2. Grind the sample under liquid nitrogen to a fine powder using a mortar and pestle.

Step 2 Lysis

1. Add 500 µl of the Buffer PL and 0.5 µl of the RNase A (50 mg/ml) to the sample in the mortar and grind the sample until it is completely dissolved.
2. Transfer the dissolved sample to a 1.5 ml microcentrifuge tube.
3. Incubate at 75°C for 30 minutes. (invert the tube every 10 minutes)
4. Centrifuge at 14-16,000 x g for 5 minutes.
5. Transfer the supernatant to a new 1.5 ml microcentrifuge tube. #Pre-heat the Buffer BE to 75°C for Step 5 DNA Elution

Step 3 DNA Binding

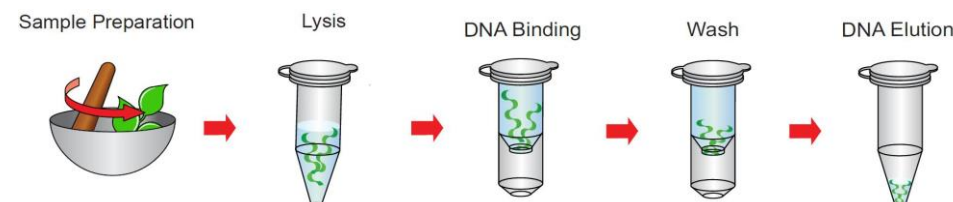
1. Add the same volume of the isopropanol to the clear supernatant from the previous step and vortex immediately for 5 seconds (eg. add 350 µl Isopropanol to the 350 µl supernatant)
2. Place a Column PC in a 2 ml Collection Tube.
3. Transfer the mixture to the Column PC.
4. Centrifuge at 14,000 x g for 30 seconds.
5. Discard the flow-through and place the Column PC back in the same Collection Tube.

Step 4 Wash

1. Add 400 µl of the Buffer W1 into the Column PC.
2. Centrifuge at 14,000 x g for 30 seconds.
3. Discard the flow-through and place the Column PC back into the same Collection tube.
4. Add 600 µl of the Buffer W2 (Ethanol added) into the Column PC.
5. Centrifuge at 14,000 x g for 30 seconds.
6. Discard the flow-through and place the Column PC back into the same Collection tube.
7. Centrifuge at 14,000 x g again for 2 minutes to remove the residual Buffer W2.

Step 5 DNA Elution

1. Transfer the dried Column PC to a new 1.5 ml microcentrifuge tube.
2. Add 50-100 µl of the Pre-Heated Buffer RE or TE into the center of the column matrix.
3. Let stand at 75°C for 3 minutes.
4. Centrifuge for 2 minutes at 14,000 x g to elute the purified DNA.



Troubleshooting

Refer to the table below to troubleshoot problems that you may encounter when purifying the genomic DNA with the kit.

Problem	Cause	Solution
Low yield of DNA	Incomplete lysed sample	Use the appropriate method for the lysate preparation based on the amount of the starting materials.
		Increase the digestion time.
		Make sure that the tissue is completely immersed in the Buffer PL.
	Ethanol not added to Buffer W2	Add 60 ml of the ethanol (96–100%) to Buffer W2, and shake before use.
	Incorrect elution conditions	Perform incubation at 75°C with Buffer RE before centrifugation. To recover more DNA perform a second elution step.
	Poor quality of starting material	Be sure to use fresh sample and process immediately after collection or freeze the sample at -80°C or in liquid nitrogen. The yield and quality of DNA isolated depends on the type and age of the starting material.
DNA degrade	Sample not fresh	Avoid repeated freeze / thaw cycles of the sample.
		Use a new sample for the DNA isolation.
		Perform the extraction of the fresh material when possible.
	DNase contaminant	Use the fresh TAE or TBE electrophoresis buffer.
		Maintain a sterile work environment to avoid contamination from DNases.
	Incomplete lysed sample	Avoid extensive pipetting to facilitate lysis/ homogenization or vortexing to prevent any DNA damage.
Presence of RNA	RNA contamination	Perform RNase A digestion step during Step Lysis.
Inhibition of downstream	Presence of ethanol in purified DNA	Discard the ethanol of Buffer W2 flow- through from the collection tube. Place the spin cartridge into the collection tube and centrifuge the spin cartridge at maximum speed for 2- 3 minutes to completely dry the cartridge.

Caution

- Buffers W1 contain irritants. Wear gloves when handling these buffers.
- Add 60 ml of the ethanol to the Buffer W2 before use.
- During operation, always wear a lab coat, disposable gloves, and protective equipment.
- Check buffers before use for salt precipitation. Redissolve any precipitate by warming to 37°C.
- Research Use Only. Not intended for any animal or human therapeutic or diagnostic uses.