

ONESpin™ Rapid Plant DNA Extraction Kit

(ONE-PLDNA-50)

Principle:

The **ONESpin™ Rapid Plant DNA Extraction Kit** utilizes silica spin column–based purification technology for rapid isolation of high-quality genomic DNA from plant tissues.

Plant cells are disrupted using mechanical and chemical lysis to release nucleic acids. The lysate is clarified by centrifugation to remove insoluble debris such as cell wall fragments and particulates.

Under optimized chaotropic salt conditions, genomic DNA selectively binds to the silica membrane of the spin column, while proteins, polysaccharides, polyphenols, secondary metabolites, and other contaminants pass through.

The bound DNA is washed sequentially using wash buffers to remove residual impurities and inhibitors. A final dry spin ensures complete removal of ethanol and wash buffer residues.

Purified DNA is then eluted under low-salt conditions into a clean microcentrifuge tube. The isolated DNA is suitable for direct application in PCR, qPCR, restriction digestion, and next-generation sequencing workflows.

Kit Contents:

Components	Quantity (50 Reactions)
PL Buffer	40 mL
BB Buffer	30 mL
Spin Columns	50
BW Buffer (Concentrate)	15 mL
BE Buffer	3 mL

Material's/Equipment's required but not Supplied:

- Ethanol (96–100%), Molecular Grade
- Micropipettes and sterile tips
- 1.5 mL microcentrifuge tubes
- Heating block or Water bath
- Vortex mixer
- Microcentrifuge

Storage Conditions:

All components of the **ONESpin™ Rapid Plant DNA Extraction Kit** should be stored at room temperature (~25°C). The kit is stable until the expiration date printed on the product label when stored under recommended conditions.

Preparation of Buffers**Preparation of BW Buffer**

BW Buffer is supplied as a concentrate and must be diluted with ethanol before use.

Component	Volume for 50 Reactions
BW Buffer (Concentrate)	15 mL
Ethanol (96-100%)	45 mL
Final Volume	60 mL

Mix thoroughly and store at room temperature.

Protocol:

1. Add 700 μ L of PL buffer to 50 to 100 mg of plant tissue and homogenize well.
2. Incubate at 65 °C for 15 minutes.
3. Invert the tube several times during incubation to mix the sample.
4. Centrifuge at 6,000 rpm for 10 minutes and transfer 500 μ L of lysate to a new 1.5 mL tube.
5. Add 400 μ L of BB buffer and vortex well.
6. Transfer the mixture to a spin column placed in a collection tube.
7. Centrifuge at 8,000 rpm for 1 minute. Discard the flow-through and place the column back into the collection tube.
8. If the sample volume exceeds the column capacity, repeat step 6–7 until all the lysate has been processed.
9. Add 500 μ L of BW buffer to the spin column and centrifuge at 10,000 rpm for 1 minute. Discard the flow-through.
10. Repeat the wash step once more with BW buffer.
11. Perform a dry spin at 10,000 rpm for 2 minutes to remove residual buffer.
12. Transfer the spin column to a new 1.5 mL microcentrifuge tube.
13. Add 50 μ L of BE buffer directly to the center of the membrane.
14. Incubate at room temperature for 5 minutes.
15. Centrifuge at 8,000 rpm for 1 minute to elute DNA. Store the eluted DNA at -20 °C.

Troubleshooting Guide:

Problem Observed	Possible Cause	Recommended Solution
Low DNA yield	Incomplete tissue lysis	Ensure proper homogenization. Grind thoroughly (especially tough tissues like leaves, seeds). Extend incubation to 25–30 min if required.
	Insufficient PL buffer penetration	Increase mixing and ensure tissue is fully submerged in buffer.
	Inefficient elution	Pre-warm BE buffer (60°C) and increase incubation time to 10 minutes before centrifugation.
No DNA or very low concentration	Ethanol not added to BW buffer	Verify BW buffer was prepared correctly with ethanol.
	Loss during lysate transfer	Carefully transfer supernatant without disturbing pellet.
	Column overloading	Reduce plant input (use 50–80 mg for high polysaccharide tissues).
Column clogging / slow flow-through	High debris content	Perform additional centrifugation (Step 4) to improve clarification.
	High polysaccharides or fibers	Reduce input material or dilute lysate before loading.
DNA degradation	Nuclease activity	Use fresh samples or store samples properly (liquid nitrogen / -80°C). Work quickly.
	Overheating during incubation	Maintain correct incubation temperature (65°C). Avoid prolonged incubation.
Brown/colored DNA (impure DNA)	Presence of polyphenols	Add extra wash step with BW buffer.
	Oxidation of plant compounds	Use fresh tissue; process immediately after grinding.
PCR inhibition	Carryover of inhibitors (polyphenols, polysaccharides)	Perform additional wash step. Dilute DNA (1:10) before PCR.
	Incomplete washing	Ensure both BW washes are done properly.
Residual ethanol contamination	Incomplete drying of column	Increase dry spin time (Step 11) to 3–5 minutes.
Inconsistent results between samples	Variation in plant type	Optimize input amount and lysis conditions depending on tissue (leaf, root, seed).