WizPrep™ Plant DNA Mini Kit

- W71650-50 **50 prep**
- W71650-150 **150 prep**

Description

The WizPrep™ Plant DNA Mini Kit provides a fast and simple method to isolate total DNA (genomic DNA, mitochondrial and chloroplast) from plant tissue and cells including leaves, stems, buds, flowers, fruit, seeds etc.

The WizPrep™ Plant DNA Mini Kit uses silica-membrane technology to eliminate the cumbersome steps associated with loose resins or slurries. The kit is ready for use and can purify the genomic DNA from a wide variety of plant species and tissues, and the whole process is completed in less than 60 minutes.

Purified DNA is suitable for PCR, restriction endonuclease digestion and Southern Hybridization.

Kit Contents

Contents	50 prep	150 prep	Storage
GP1 Buffer	25 ml	80 ml	Room temp.
GP2 Buffer	6 ml	20 ml	Room temp.
GP3 Buffer	35 ml	110 ml	Room temp.
PW1 Buffer (concentrate)	12 ml	40 ml	Room temp.
PW2 Buffer (concentrate)	14 ml	44 ml	Room temp.
Elution Buffer	5 ml	20 ml	Room temp.
RNase A (lyophilized)**	3 mg	9 mg	4℃
Spin Columns*	50	150	Room temp.
Filter Columns*	50	150	Room temp.
Collection Tubes (2.0ml)	100	300	Room temp.
Instruction Manual	1	1	Room temp.

* All Spin Columns are sterilized by electron beam.

** After receiving the RNase A, please store at 4℃.
After dissolved in distilled water, store the RNase A solution at 4℃ for up to 3 months. For longer storage (up to 1 year), the RNase A solution should be divided into small aliquots and stored at -20℃.

Reagents and equipment to be supplied by user

- 96~100% ethanol (to prepare PW1 and PW2 Buffer)
- 1.5 mL microcentrifuge tubes
- Sterile RNase-free pipette tips and Manual pipettors
- Centrifuge for microcentrifuge tubes
- Equipment for sample disruption and homogenization
- Personal protection equipment (lab coat, gloves, goggles)

Kit specifications

Parameter	Characteristics
Format	Silca-membrane spin column
Sample materials	< 100 mg tissue
Fragment size	> 200 nt
Typical yield	1~30 ug from 100 mg plant material
$A_{260/280}$	1.7 ~ 1.9
Elution volume	50 µl
Preparation time	<60 minutes
Binding capacity	200 ug

Quality Control Analysis

The kit was qualified by isolating genomicDNA from 100 mg of plant tissue following the protocols outlined in the manual. The purified genomic DNA has an A260/280 ratio between 1.7 and 1.9.

Quality Authorized by : Jamie Ahn

Protocol

Before starting :

1) Add 12 (40) ml of 100% ethanol to the 12 (40) ml of PW1 Buffer.
2) Add 21 (66) ml of 100% ethanol to the 14 (44) ml of PW2 Buffer.
3) Dissolve RNase A powder in 300 µl distilled water.

Step 1 : Prepare plant tissue

- Transfer ≤100 mg of plant tissue or $5x10^6$ plant cells into a mortar that contains an appropriate amount of liquid nitrogen to cover the sample.
- Grind the sample into a fine powder using a pestle in liquid nitrogen.

Note: If stored frozen samples are used, do not allow the samples to thaw before transferring to the liquid nitrogen. Allow the liquid nitrogen to evaporate, without allowing the tissue to thaw.

Step 2 : Lysis step

- Add **400µl of GP1 Buffer** and **5µl of RNase A** (10 mg/ml) into the sample tube and mix by vortexing. Do not mix GP1 Buffer and RNase A before use.
- Incubate at 65℃ for 10 minutes. During incubation, invert the tube every 5 minutes.
- Add **100µl of GP2 Buffer** and mix by vortexing.
- Incubate on ice for 3 minutes.
- Place a Filter Column in a 2ml Collection Tube.
- Transfer the lysate (500µl) mix to Filter column and centrifuge for 1 min. at 13,000 rpm.
- Discard the Filter Column and carefully transfer supernatant (400µl) in Collection Tube to a new microcentrifuge tube (provided by user).

Step 3 : Binding step

- Add **1.5 volume (600µl) of GP3 Buffer** to filterate and mix by invert for 5 times.
- Connect Spin Column to 2.0 ml Collection tube.
- Apply the mixture to the Spin Column and centrifuge for 2 min. at 13,000 rpm.
- Discard the flow-through and re-connect with Spin Column.

Step 4 : Wash step

- Add **400µl of PW1 Buffer** to the Spin Column and centrifuge for 30 sec. at 13,000 rpm.
- Discard the flow-through and re-connect with the Spin Column.
- Add **600µl of PW2 Buffer** (ethanol added) in the center of Spin Column matrix and centrifuge for 30 sec. at 13,000 rpm.
- Discard the flow-through and re-connect the Spin Column and centrifuge for 3 min. at 13,000 rpm.

Step 5 : Elution step

- Connect the Spin Column and new 1.5 ml tube.
- Add **50µl of Elution Buffer** and incubate at R/T for 1 min.
- Centrifuge for 1 min. at 13,000 rpm.
- Discard the Spin Column and eluted purified DNA for use next step.
- The purified DNA sample may be stored at −20°C for a few days. It is recommended that samples be placed at −70°C for long term storage.

Quick Protocol

Homogenization

GP1 Buffer (400µl)
RNase A (5µl)
65℃ / 10 min.

GP2 Buffer (100µl)
Ice / 3 min.

Filter Column

Filterate (400µl)
GP3 Buffer (600µl)

PW1 Buffer (400µl)

PW2 Buffer (600µl)

Elution Buffer (50µl)

RUO Research Use Only

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Troubleshooting

Problem	Possible cause and solution
Column clogged	Sample overloading • Reduce sample volume or separate into multiple tubes. Precipitate was formed at DNA Binding Step • Reduce the sample material. • Before loading the column, break up precipitate in ethanol-added lysate
Low yield	Incorrect DNA Elution Step • Ensure that Elution Buffer was added and absorbed to the center of Spin Column matrix. Incomplete DNA Elution • Elute twice to increase yield
Eluted DNA does not perform well in downstream applications	Residual ethanol contamination • Following the washing step, dry Spin Column with additional centrifugation at full speed for 5minutes or incubation at 60℃ for 5 minutes. RNA contamination • Perform Optional RNA Degradation Step. Protein contamination • Reduce the sample amount. • After DNA Binding Step, apply 400µl PW1 Buffer to wash Spin Column and centrifuge at 13,000 rpm for 30 seconds. Proceed with Washing Step of PW2 Buffer. Genomic DNA was degraded • Use fresh sample, long storage may result in fragmentation of genomic DNA.

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