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VER.:07A

VIOGENE

User Bulletin

Plant Genomic DNA Extraction Miniprep System

Isolation of genomic DNA from 100 mg plant material or 1×10^8 cells.

Downstream Application

- * Restriction digestion
- * Southern Blotting
- * RAPD, RFLP
- * PCR, Real-Time PCR

Product Contents

Cat. No	GPG1001	GPG1002
Preps	50	250
PX1 Buffer	24ml	120ml
PX2 Buffer	8ml	40ml
PX3 Buffer	18ml	90ml
RNase A	20mg	110mg
WS Buffer	15ml	45ml x 2
Plant Genomic DNA Mini Column	50	250
Shearing Tube	50	250
Collection Tube	100	500
Protocol	1	1

All buffers need to be mixed well before use.

Shipping & Storage

Viogene Plant Genomic DNA Extraction Miniprep is shipping and storage at ambient temperature up to 12 months.

If precipitate form by freezing temperature on any buffer, warm up at 37

to redissolve.

Protocol

❖ **Please read the following notes before starting the procedures.**

Important Notes

- All centrifugation should be done at room temperature.
- Preheat a water bath to 65 °C.
- Preheat TE or ddH₂O to 65 °C for DNA elution.
- PX1 buffer may form unclear upon storage. This does not affect the procedure and efficiency.
- PX1 Buffer may form a precipitate, warm at 65 °C to redissolve.

For GPG1001

- Add 200 µl of ddH₂O to the RNase A powder tube, vortex to dissolve and store at 4 °C.
- Add 60 ml of ethanol (98~100 %) to the WS Buffer bottle when first open the bottle.

For GPG1002

- Add 1100 µl of ddH₂O to the RNase A powder tube, vortex to dissolve and store at 4 °C.
- Add 180 ml of ethanol (98~100 %) to the WS Buffer bottle when first open the bottle.

- 1. Grind 100 mg (or less) plant sample under liquid nitrogen to fine powder and transfer to a new tube.**

Do not allow the sample to thaw, and continue immediately to step 2.

- 2. Add 400 μ l of PX1 Buffer and 4 μ l of RNase A solution (100 mg/ml) to the tissue powder and vortex vigorously, then incubate the mixture at 65 °C for 10 minutes.**

Do not mix PX1 Buffer and RNase A prior to use. Invert 2-3 times during 65 °C incubation.

- 3. Add 130 μ l of PX2 Buffer to the lysate, vortex, and incubate on ice for 5 minutes.**

- 4. Apply lysate to the Shearing Tube sitting in a Collection Tube and centrifuge at full speed for 2 minutes. Transfer flow-through sample from the Collection Tube to a new tube.**

Avoid pipetting any debris or pellet in the collection tube.

- 5. Add 0.5 volume of PX3 Buffer and 1 volume of 98-100% ethanol to the clear lysate and invert the tube 3 ~ 5 times.**

For example: If 450 μ l clear lysate collected, add 225 μ l PX3 Buffer and 450 μ l ethanol.

- 6. Apply 650 μ l of the ethanol added sample (including any precipitate) from step 5 to a Plant Genomic DNA Mini Column sitting in a Collection Tube, close the cap, centrifuge at 8,000 x g (10,000 rpm) for 1 minute, and discard the filtrate.**

If the solution remains above the membrane, centrifuge again at 13,000 rpm.

- 7. Repeat step 6 for rest of the sample.**

- 8. Wash the column twice with 0.7 ml of WS Buffer by centrifuging at full speed (13,000 rpm or 10,000 x g) for 30 ~ 60 seconds and discard the filtrate.**

Add ethanol (see [Important Notes](#)) to the WS Buffer bottle when first open the bottle.

- 9. Centrifuge at full speed for 3 minutes to remove traces of WS Buffer.**

- 10. Transfer the column to a new 1.5 ml tube, add 200 μ l of 65 °C TE or ddH₂O, and centrifuge at full speed for 1 minute to elute DNA.**

- 11. Store DNA at -20 °C.**