Genomic DNA Isolation Kit (Plant)

Cat. No.:	YGL100	YGL500
Product Name:	Genomic DNA Isolation Kit (Plant)	
Reactions:	100	500
Sample:	Fresh or dry plant tissue	
Yield:	Up to 80ug for 100mg of fresh plant tissue	
Format:	Reagent	
Operation:	Centrifuge	
Operation Time:	Within 90 Minutes	

Description

Genomic DNA Isolation Kit (Plant) enables 3-steps operations to isolate total DNA (including genomic, mitochondrial and chloroplast DNA) from plant tissue and cells. Samples are initially disrupted by grinding in liquid nitrogen, followed by lysis treatment with RNase A. The unique GL Buffer is able to lyse most common plant samples and also samples high in polysaccharides. DNA phenol extraction is not required and the entire procedure can be completed in 1.5 hours. The extracted total DNA is ready for use in PCR, Real-time PCR, Southern Blotting, Mapping and RFLP.

Features

1. Convenient, scalable purification procedure.
2. Reproducible recoveries for constant results.
3. Complete removal of all contaminants for reliable downstream applications.

Contents

ITEM	YGL100	YGL500
GL Buffer	100ml	500ml
RNase A (50mg/ml)	50ul	250ul

*If GL Buffer contains sediment, incubate at 65ºC for 10 minutes to dissolve.
**Add RNase A to GL Buffer prior to use.

Applications

The extracted total DNA is ready for use in PCR, Real-time PCR, Southern Blotting, Mapping and RFLP.

Quality Control

The quality of Genomic DNA Isolation Kit (Plant) is tested on a lot-to-lot basis by isolation of Genomic DNA from 100 mg of fresh plant leaf. Genomic DNA is quantified with a spectrophotometer and the yield of Genomic DNA is more than 20 ug. The purified DNA is checked by electrophoresis.

Caution

The components contain irritants. During operation, always wear a lab coat, disposable gloves, and protective goggles.

Protocol

Additional Requirements:
Microcentrifuge tubes, Absolute Ethanol for preparing 70% Ethanol in water, Chloroform, Isopropanol, Mortar and Pestle.

Optional requirements:
If a larger sample volume is required, scale the GL Buffer proportionately.

Tissue Dissociation	★Cut off 100 mg of fresh plant issue or 50 mg of dry plant tissue. ★Grind the sample under liquid nitrogen to a fine powder using a mortar and pestle.
Step 1 Lysis	★Add **1 ml of GL Buffer and 0.5 µl of RNase A** to the sample in the mortar and grind the sample until it is completely dissolved. ★Transfer the dissolved sample to a 1.5 ml microcentrifuge tube. ★Incubate at 65ºC for 50 minutes, followed by full speed centrifugation for 10 minutes.
Step 2 Isolation	**Standard Samples** ★Transfer the supernatant to a new 1.5 ml microcentrifuge tube containing 600 µl of chloroform. ★Shake vigorously and then centrifuge at full speed for 10 minutes. ★Carefully remove the upper layer and transfer it to a new 1.5 ml microcentrifuge tube.
	High Polysaccharide Samples ★Transfer the supernatant to a new 1.5 ml microcentrifuge tube and add a 1/10 volume of GL Buffer and 600 µl of chloroform. ★Shake vigorously and then centrifuge at full speed for 10 minutes. ★Carefully remove the upper layer and transfer it to a new 1.5 ml microcentrifuge tube (repeat the Isolation Step until the interphase becomes clear).

Step 3 Nucleic Acid Precipitation	★Add 800 µl of isopropanol to the 1.5 ml microcentrifuge tube (containing the upper layer) from step 2. ★Mix the sample by inverting gently and let stand for 5 minutes at room temperature (DNA precipitation can be increased with extended standing time). ★Centrifuge at full speed for 20 minutes. ★Discard the supernatant and wash the pellet with 1 ml of 70% ethanol and air dry. ★Resuspend the pellets in 50-200 µl of 1 x TE buffer or water. ★Incubate for 10 minutes at 60ºC to soften the pellet. ★Dissolve the pellet completely with pipetting. ★Centrifuge at full speed for 10 minutes. ★Carefully transfer the supernatant to a new 1.5 ml microcentrifuge tube.

Troubleshooting

Problem	Possible Reasons/Solution
Incomplete Lysis	**Too much sample was used** ★Reduce sample volume or separate into multiple tubes and grind the sample completely
Low Yield	**Precipitate was formed at Step 3 Nucleic Acid Precipitation** ★Reduce the sample material. ★Increase standing time to improve DNA precipitation.