G2P Gene to Protein Pvt. Ltd.

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800 GENOME, 800 GENETIC

G2P Plant DNA Purification Kit (Spin Column Based)

Cat # ☐ **PUR16-50** ☐ **PUR16-250** ☐ **PUR16-50-S**
Pack Size: . ☐ **50 Preps ,** ☐ **250 Preps,** ☐ **5 Preps**
Storage: Room Temperature*

Introduction:

The G2P Plant DNA purification kit is a state-of-the-art solution that addresses the challenges of DNA extraction from plant tissue, allowing for the recovery of high-quality DNA from freshly growing plant leaves. With advanced features, this kit offers improved removal of inhibitors and increased quantities of plant and plant pathogen DNA, resulting in high-performance outcomes in downstream applications. The silica-based DNA isolation process is both rapid and straightforward, utilizing a spin column configuration. Depending on the type of sample utilized, typical yields range from 3 to 15µg of high-quality DNA.

Kit contents: * Store at 4°C

S.No	Components	PUR16-50	PUR16-250	PUR16-50-S
1	Buffer PDC	30mL	150mL	3mL
2	Rnase A(10mg/mL)*	500 µl	2.5mL	50 µl
4	Buffer D (Wash Buffer)	10mL	50mL	2mL
5	Buffer E (Elution Buffer)	5mL	25mL	500 µl
6	Spin Columns and Collection Tubes	50	250	5

Reagents/material required but Not Provided

- Chloroform:Isoamyl alcohol (24:1)
- 100% Ethanol
- Plastic Pestle/Tissue Lyser: For Mechanical lysis of the plant tissue

Protocol

Note: Add 40 ml (1:4) of absolute ethanol in Buffer D (Wash buffer) before starting the protocol

1. Take ~50-80mg fresh growing leaf/pulp/soft stem tissue into a 1.5ml microcentrifuge tube
2. Add 600 µl of Buffer PDC and homogenize well using plastic pestle
   Note: *A good homogenization of the plant tissue is must to get a good quality and quantity of DNA. We highly recommend to use plastic pestle coupled with mechanical homogenizer/tissue lyser. If homogenizer/lyser is not available, use the liquid nitrogen base tissue pulverization method.*
3. Incubate at 55°C for 30 min
   Note: If the tissue is hard homogenize again after 10 min of incubation
4. Add 10 µl of RNase A and incubate at 37°C for 30 min
5. Add 600 µl of Chloroform:Isoamyl alcohol (24:1) in the above mix and invert mix
6. Centrifuge at 12000 x g for 10 min at 4°C
7. Transfer the upper aqueous layer into a fresh 1.5ml microcentrifuge tube
8. Repeat step 5-7 once again (Optional step for better quality)
9. Add 1 volume 100% chilled isopropanol invert mix and load it on the column and wait for 2 min
10. Centrifuge at 12000 x g for 5 min at 4°C
    Note*: If the entire volume has not passed through the column then increase the centrifugation time*
11. Discard the flow through which is collected in the collection tube
12. Add 500 µl of buffer D on the spin column and centrifuge at 12000 x g for 2 min at 4°C
13. Discard the flow through which is collected in the collection tube
14. Repeat step 12-13 once again
15. Dry spin the spin column by centrifuge at 12000 x g for 5 min at 4°C
16. Put the spin column into a fresh 1.5 ml microcentrifuge tube and add 50 µl of buffer E
17. Centrifuge at 10000 x g for 2 min to collect the pure DNA

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Graphical Protocol

50-80 mg

600 µl PDC

Homogenization

Incubate at 55°C
For 30 min

Add 10 µl RNase A
and incubate at 37°C
For 30 min

Add 600 µl
Chloroform:Isoamyl alcohol
(24:1)

Centrifuge 12000 x g
for 10 min

Aqueous
Phase

Intermediate
Phase

Organic
Phase

Carefully withdraw
the aqueous phase

Transfer to a
fresh vial

Again add 600 µl
chloroform:isoamyl alcohol (24:1)

Invert mix and
centrifuge 12000 x g for 10 min

Carefully withdraw
the aqueous phase

Transfer to a
fresh vial

Add 1 Volume
100% chilled Isopropanol
and invert mix

Load it on the
spin column

Centrifuge 12000 x g
for 5 min

Flow
through

Discard the
flow through

Add 500 µl
buffer D

Centrifuge 12000 x g
for 2 min

Flow
through

Discard the
flow through

Add 500 µl
buffer D

Discard the
flow through

Dry spin
12000 x g
for 5 min

Transfer the
column to
a fresh vial

Add 50 µl
buffer E

Centrifuge 10000 x g
for 2 min

Pure DNA

Result

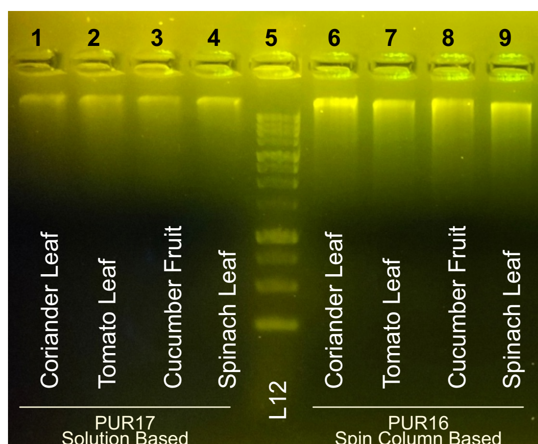

Fig: Various plant genomic DNA was isolated using PUR17 (lane 1-4) and PUR16 (lane 6-9). 100ng of DNA sample from each plant was loaded on 0.8% agarose gel.