Exgene™ Plant SV is specialized for extracting high-quality genomic DNA from various plant leaves

Experimental Conditions

Materials Required

- Exgene™ Plant SV, mini (117-101, 100 preps)
- Grinder : TissueLyser II (85300, Supplier : Q) or another grinding devices with equivalent performance
- Liquid nitrogen (LN_2)
- Absolute ethanol (EtOH, C_2H_6O, CAS No. : 64-17-5, ≥99.0%)
- 1.5 ml or 2.0 ml microcentrifuge tube
- Vortex mixer
- Centrifuge (Max. speed 14,000 rpm or ≥10,000 x g)
- Pipette & sterile pipette tips
- Suitable protector (ex. lab coat, disposable gloves, goggles, etc.)
- 65˚C water bath or heating block (for incubation)
- Ice (for incubation)

Sample Information

- Sample type : Plant leaf

Garlic L. | Maize L. | Cucumber L. | Persimmon L.
Forsythia L. | Tomato L. | Pumpkin L. | Red giant mustard L.
Jujube L. | Lettuce L.

- Sampling : after collecting fresh leaves, put them in a bag and seal it.
- How to store : store at -70˚C temperature in a deep freezer
- Grinding : TissueLyser II (85300, Supplier : Q) or another devices
- Extraction conditions
  - Sample amount : 100 mg
  - Elution volume : 50 µl

Protocol

Before Starting

- Before using for the first time, add absolute ethanol (ACS grade or better) into Buffer BD and Buffer CW as indicated on the bottle.
- Buffer PL may precipitate upon storage at cold ambient temperature. If so, dissolve it in 65˚C water bath.

Sample Preparation

1. Freeze the leaf samples using LN_2.
2. Grind the frozen samples using Tissuelyser II (85300, Supplier : Q) or another equipment.
3. Measure the weight of 100 mg and transfer leaf samples to 1.5 or 2.0 ml microcentrifuge tube.
4. The next step is according to **Exgene™ Plant SV mini protocol**.

Exgene™ Plant SV mini Brief Protocol

*** For more details and methods, please refer to the handbook of Exgene™ Plant SV mini**

Lysis

1. Add 400 µl Buffer PL and 4 µl of RNase A solution into the sample tube and vortex vigorously.
2. Incubate for 10~15 min at 65˚C and mix 2~3 times during incubation.

Filtration

3. Add 140 µl Buffer PD to the lysate. Vortex and incubate for 5 min on ice.
4. Transfer the mixture to EzSep™ Filter and centrifuge at 10,000 x g for 2 min.

Binding

5. Transfer 450 µl of the pass-through to a new 1.5 ml microcentrifuge tube.
6. Add 1.5 volumes of Buffer BD and Mix immediately.
7. Transfer 700 µl of the mixture to Column Type G and centrifuge at 10,000 x g for 30 sec.
8. Transfer the all of remaining mixture to Column Type G and centrifuge at 10,000 x g for 30 sec.

Washing

9. Add 700 µl Buffer CW to Column Type G and centrifuge at 10,000 x g for 30 sec.
10. Add 300 µl Buffer CW to Column Type G and centrifuge at 10,000 x g for 2 min.
11. Transfer Column Type G to a new 1.5 ml microcentrifuge tube.

Elution

12. Add 50 µl Buffer AE to Column Type G.
    Incubate for 5 min at RT and centrifuge at 10,000 x g for 1 min.

Table 1. Brief Protocol of GeneAll® Exgene™ Plant SV mini kit for gDNA purification from various plant samples

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Result

#	Sample	Conc. (ng/µl)	Yield (µg)	$A_{260/280}$	$A_{260/230}$
1	Garlic L.	273.7	**13.69**	1.85	2.55
2	Maize L.	90.7	**4.54**	1.79	2.91
3	Cucumber L.	132.8	**6.64**	1.85	2.63
4	Persimmon L.	388.1	**19.40**	1.86	2.18
5	Forsythia L.	142.4	**7.12**	1.82	2.22
6	Tomato L.	175.0	**8.75**	1.84	2.30
7	Pumpkin L.	202.7	**10.14**	1.80	2.19
8	Red giant mustard L.	181.5	**9.07**	1.83	2.55
9	Jujube L.	412.2	**20.61**	1.87	2.09
10	Lettuce L.	272.7	**13.64**	1.84	2.28

Table 2. The concentration, yield and purity (mean) of gDNA extracted from 100 mg of various leaf samples.

※ Absorbance measurement instrument : NanoDrop™ 2000/2000c (ND-2000, Supplier : T)

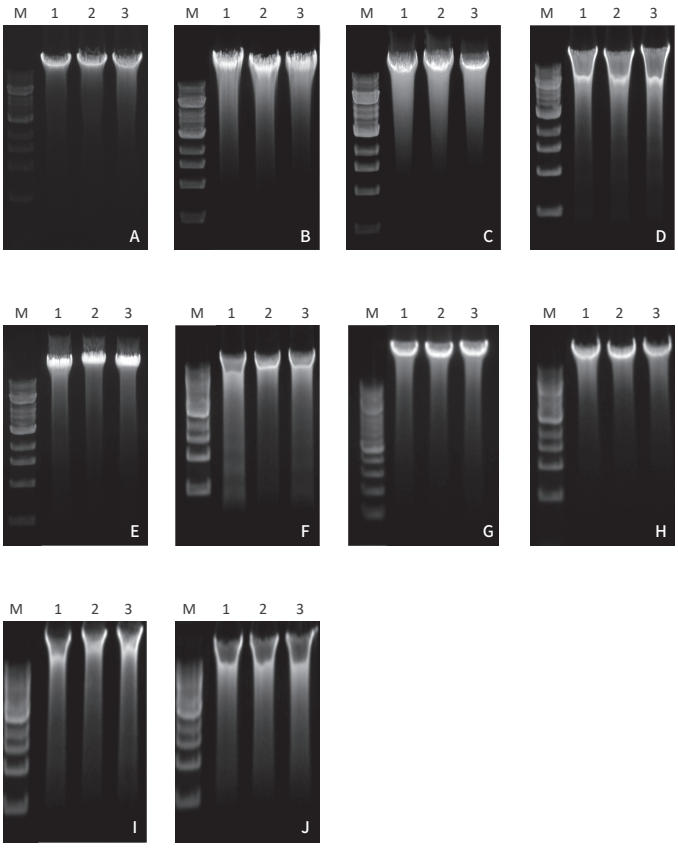

Figure 1. The results of electrophoresis of gDNA from 100 mg of various plant leaf samples.
M : GENESTA™ 1 kb DNA Ladder with 5X loading dye (GA-100, GeneAll®, 1 µl loading)
A : garlic leaf, **B** : maize leaf, **C** : cucumber leaf, **D** : persimmon leaf, **E** : forsythia leaf, **F** : tomato leaf, **G** : pumpkin leaf, **H** : red giant mustard leaf, **I** : jujube leaf, **J** : lettuce leaf

※ Electrophoresis conditions : 1.0% agarose gel, 150 V, 20 min, 5 µl loading