

Description:

Plant Genomic DNA Extraction Mini Kit is an efficient and easy-to-use tool and specially designed for genomic DNA extraction from plant tissue or cultured plant cells. The procedure includes plant tissue lysis, optimized enzyme treatment to remove RNA from the sample mixture and filter columns removing unwanted particles. Silica-based FSPG Column and FSPG3 Buffer are specially designed for plant genomic DNA extraction that enables optimal performance. This kit provides an easy and efficient method to purify plant genomic DNA.

Kit Contents:

	DE-021s (4 preps_sample)	DE-022 (100 preps)
FSPG1 Buffer	2.0 ml	55 ml
FSPG2 Buffer	1.0 ml	15 ml
FSPG3 Buffer * (concentrate)	1.5 ml	30 ml
Wash Buffer W1 * (concentrate)	1.3 ml	26 ml
Wash Buffer W2 * (concentrate)	1.5 ml	30 ml
Elution Buffer	1.5 ml	30 ml
RNase A (lyophilized)	1.5 mg	43 mg
Filter Column	4 pcs	100 pcs
FSPG Column	4 pcs	100 pcs
Collection Tube	8 pcs	200 pcs

* Preparation of FSPG3 Buffer and Wash Buffers for first use:

Cat. No:	DE-021s (4 preps)	DE-022 (100 preps)
ethanol volume for FSPG3 Buffer	3 ml	60 ml
ethanol volume for Wash Buffer W1	1.0 ml	34 ml
ethanol volume for Wash Buffer W2	6 ml	120 ml

Specification:

Principle: Spin Column (silica membrane)

Sample: 1.Wet weight ≤ 100 mg - 2.Dry weight ≤ 20 mg

Operation time: 30~ 60 min

Expected DNA Yield: 5~40 ug

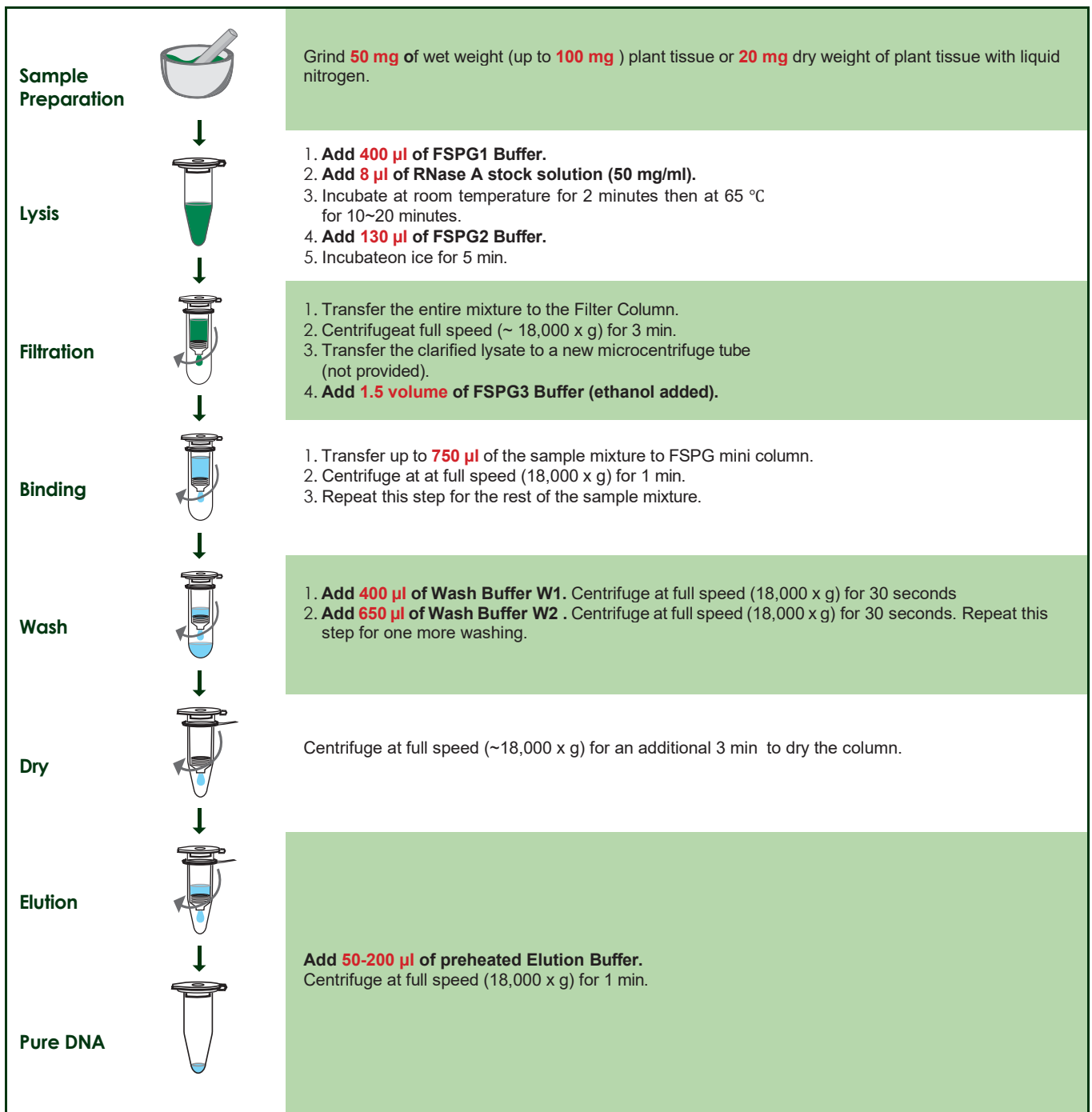
Minimum Elution Volume: 50ul

Column suitability: Centrifugation/ Vacuum

Important Notes:

1. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
2. Check FSPG1 Buffer before use, Warm FSPG1 Buffer at 60°C for 5 minutes if any precipitate formed.
3. Preheat dry baths or water baths to 65°C before the operation.
4. **Add required ethanol** (96-100%) to FSPG3 Buffer, to Wash Buffer W1 and to Wash Buffer W2 before use.
5. Store RNase A at -20°C upon arrival, Add sterile ddH₂O to RNase A tube to make a 50 mg/ml stock solution. Vortex and make sure that RNase A has been completely dissolved. Store the stock solution at +4°C.
6. All centrifuge steps are done at full speed (~ 18,000 x g) in a microcentrifuge.

Sample	DNA Yield (ug)
	100 mg young leaf
Arabidopsis	3~5
Rice	10 ~15
Tomato	10 ~15
Tobacco	20~25
Chinese Yam	30~60
Maize	15~20
Sweet Potato	20~30
Orchis	5~10
Campor Tree	15~20
Spinach	5~10
Bamboo	10 ~15



General Protocol:

HINT: Prepare a ice box and a 65 °C bath for step 2 and 3. Preheat Elution Buffer to 65 °C for step 13 (elution step).

- 1. Grind 50 mg of wet weight (up to 100 mg) or 20 mg dry eight plant tissue with liquid nitrogen to a fine powder and transfer to a new microcentrifuge tube (not provided).**
 - Do not allow the sample to thaw, and continue immediately to step 2.
- 2. Add 400 µl of FSPG1 Buffer and 8 µl of RNase A stock solution (50 mg/ml) to the tissue powder and vortex vigorously.**
- 3. Incubate the mixture at room temperature for 2 minutes then at 65°C for 10~20 minutes and invert 2-3 times during incubation.**
- 3. Filtration:**Add 130 µl of FSPG2 Buffer to the mixture. Vortex to mix well and incubate the mixture on ice for 5 min.
- 4.** Place a Filter Column to a Collection Tube and transfer the entire mixture from previous step to the Filter Column. Centrifuge the Filter Column at full speed (~ 18,000 x g) for 3 min.
- 5.** Transfer the clarified lysate (supernatant) from the Collection Tube to a new microcentrifuge tube (not provided). Discard used Filter Column and Collection Tube. And adjust the volume of clarified lysate.
Note! Do not aspirate any debris when transferring the clarified lysate.
- 6. Add 1.5 volume** of FSPG3 Buffer (ethanol added) to the clarified lysate and mix well by pipetting.
 - Make sure that ethanol (96~100%) has been added to FSPG3 Buffer when first use.
- 7.** Place a FSPG Column to a new Collection Tube and transfer **up to 750 µl** of the sample mixture from previous step carefully to the FSPG Column.
- 8.** Centrifuge at full speed (18,000 x g rpm) for 1 min. Discard the flow-through and place the FSPG Column back to the Collection Tube.

Repeat step 7 for the rest of the sample mixture.

- 9. Add 400 µl** of Wash Buffer W1 (ethanol added) to the FSPG Column. Centrifuge at full speed (18,000 x g) for 30 seconds. Discard the flow-through and place the FSPG Column back to the Collection Tube.
--Make sure that ethanol (96~100%) has been added into Wash Buffer W1 when first use.

- 10. Add 650 µl** of Wash Buffer W2 (ethanol added) to FSPG Column. Centrifuge at full speed (18,000 x g) for 30 seconds. Discard the flow-through and place the FSPG Column back to the Collection Tube.
--Make sure that ethanol (96~100%) has been added into Wash Buffer W2 when first use.
Repeat step 10 for one more washing.

- 11.** Centrifuge at full speed (18,000 x g) for an additional 3 min to dry the FSPG column completely.
--Important step! This step will avoid the residual liquid to inhibit subsequent enzymatic reactions.

- 12.**Combine the FSPG Column with a Elution Tube, Add 100 µl of preheated Elution Buffer to the membrane center of the FSPG Column. Stand the FSPG Column for 3 minute at room temperature.
--Important step! For effective elution, make sure that the elution buffer is dispensed onto the membrane center and is absorbed completely.

- 13.** Centrifuge at full speed (18,000 x g rpm) for 2min to elute purified DNA.

Troubleshooting

PROBLEM	POSSIBLE Reasons	SOLUTIONS
Incorrect preparation of FSPG3 Buffer or Wash Buffer		
Low or no yield of genomic DNA	FSPG3 Buffer is not mixed with ethanol before use	Repeat the extraction procedure with a new sample.
	Wash Buffer W1 and Wash Buffer W2 is not mixed with ethanol before use	Make sure that the correct volumes of ethanol (96- 100 %) is added into Wash Buffer W1 and Wash Buffer W2 when first open. Repeat the extraction procedure with a new sample.
	The volume or the percentage of ethanol is not correct before adding into Wash Buffer W1 and Wash Buffer W2	Make sure that the correct volumes of ethanol (96- 100 %) is added into Wash Buffer W1 and Wash Buffer W2 when first use. Repeat the extraction procedure with a new sample.
Elution of genomic DNA is not efficient		
	pH of water (ddH ₂ O) for elution is acidic	Make sure the pH of ddH ₂ O is between 7.5- 9.0. Use Elution Buffer (provided) for elution.
	Elution Buffer or ddH ₂ O is not completely absorbed by column membrane	After Elution Buffer or ddH ₂ O is added, stand the PGDE Column for 5 min before centrifugation.
Column is clogged		
	Sample is too viscous	Reduce the sample volume.
Degradation of eluted DNA		
	Sample is old	Always use fresh or well-stored sample for genomic DNA extraction.

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