

## Kit Contents:

Cat.No.	FAPGK 000 (4 preps_sample)	FAPGK 001 (50 preps)	FAPGK 001-1 (100 preps)	FAPGK 001-2 (200 preps)	FAPGK 001-3 (300 preps)
FAPG1 Buffer	2.0 ml	25 ml	55 ml	110 ml	165 ml
FAPG2 Buffer	1.0 ml	8 ml	15 ml	30 ml	45 ml
FAPG3 Buffer * (concentrate)	1.5 ml	15 ml	30 ml	60 ml	90 ml
• W1 Buffer * (concentrate)	0.8 ml	13 ml	26 ml	52 ml	78 ml
• Wash Buffer * (concentrate)	1.5 ml	15 ml	30 ml	30 ml x 2	30 ml x 3
Elution Buffer	1.5 ml	15 ml	30 ml	30 ml x 2	30 ml x 3
RNase A (lyophilized)	1.5 mg	22 mg	43 mg	43 mg x 2	43 mg x 3
Filter Column	4 pcs	50 pcs	100 pcs	200 pcs	300 pcs
FAPG Column	4 pcs	50 pcs	100 pcs	200 pcs	300 pcs
Collection Tube	8 pcs	100 pcs	200 pcs	400 pcs	600 pcs
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• The Buffer name has been changed since March 2020.

\* Preparation of FAPG1 Buffer, W1 Buffer and Wash Buffer for first use:

Cat. No:	FAPGK 000 (4 preps)	FAPGK 001 (50 preps)	FAPGK 001-1 (100 preps)	FAPGK 001-1 (200 preps)	FAPGK 001-1 (200 preps)
ethanol for FAPG3 Buffer	3 ml	30 ml	60 ml	120 ml	180 ml
ethanol for W1 Buffer	1.0 ml	17 ml	34 ml	68 ml	102 ml
ethanol for Wash Buffer	6 ml	60 ml	120 ml	120 ml	120 ml

## Specification:

Principle: spin column (silica membrane)

Sample: wet weight ≤ 100 mg

dry weight ≤ 20 mg

Operation time: < 60 min

DNA Yield: 5~40 µg

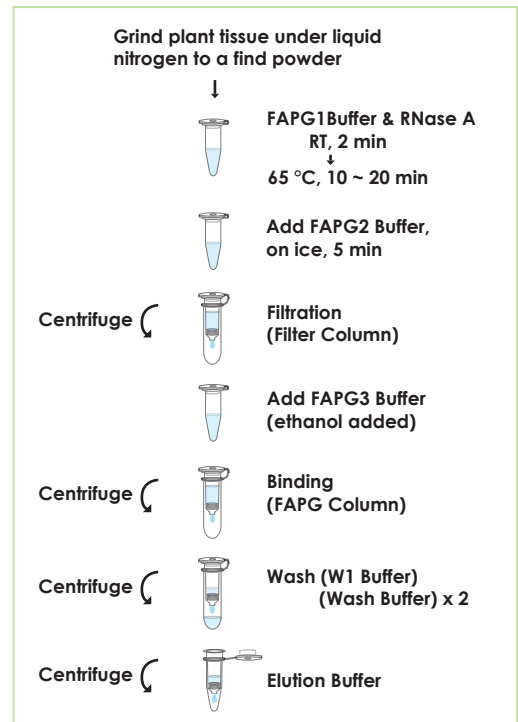
## Important Notes:

1. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
2. Check FAPG1 Buffer before use, Warm FAPG1 Buffer at 60°C for 5 min if any precipitate formd.
3. Preheat dry baths or water baths to 65°C before the operation.
4. Add required ethanol (96-100%) to FAPG3 Buffer, W1 Buffer and Wash Buffer before first use.
5. Store RNase A at -20 °C upon receipt of kit. Add sterile ddH<sub>2</sub>O 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 -20 °C

## General Protocol:

HINT: Prepare a 65 °C bath and a ice box 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) plant tissue or 20 mg dry weight of plant tissue under 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 FAPG1 Buffer and 8 µl of RNase A stock solution (50 mg/ml) to the tissue powder and vortex vigorously. 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. **Add 130 µl of FAPG2 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 measure the volume of clarified lysate.**
  - Note! Do not aspirate any debris when transferring the clarified lysate.



6. **Add 1.5 volume of FAPG3 Buffer (ethanol added) to the clarified lysate and mix well by pipetting.**
  - Make sure that ethanol (96~100%) has been added to FAPG3 Buffer when first use.
7. **Place a FAPG Column to a new Collection Tube and transfer up to 750  $\mu$ l of the sample mixture carefully to the FAPG Column. Centrifuge at full speed (18,000 x g or 14,000 rpm) for 1 minute. Discard the flow-through and place the FAPG Column back to the Collection Tube.**
8. **Repeat step 7 for the rest of the sample mixture.**
9. **Add 400  $\mu$ l of W1 Buffer (ethanol added) to the FAPG Column. Centrifuge at full speed (18,000 x g or 14,000 rpm) for 30 seconds. Discard the flow-through and place the FAPG Column back to the Collection Tube.**
  - Make sure that ethanol (96~100%) has been added into W1 Buffer when first use.
10. **Add 650  $\mu$ l of Wash Buffer (ethanol added) to FAPG Column. Centrifuge at full speed (18,000 x g or 14,000 rpm) for 30 seconds. Discard the flow-through and place the FAPG Column back to the Collection Tube.**
  - Make sure that ethanol (96~100%) has been added into Wash Buffer when first use.
11. **Repeat step 10 for one more washing.**
12. **Centrifuge at full speed (18,000 x g or 14,000 rpm) for an additional 3 minutes to dry the FAPG column completely.**
  - Important step!** This step will avoid the residual liquid to inhibit subsequent enzymatic reactions.
13. **Combine the FAPG Column with a Elution Tube, Add 50~200  $\mu$ l of preheated Elution Buffer to the membrane center of the FAPG Column. Stand the FAPG Column for 1 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.
14. **Centrifuge at full speed (18,000 x g or 14,000 rpm) for 1 minute to elute purified DNA.**

## Troubleshooting

Problems	Possible reasons	Solutions
<b>Low or no yield of genomic DNA</b>		
	Incorrect preparation of FAPG3 Buffer or Wash Buffer	
	FAPG3 Buffer is not mixed with ethanol before use	Repeat the extraction procedure with a new sample.
	W1 Buffer and Wash Buffer is not mixed with ethanol before use	Make sure that the correct volumes of ethanol (96-100 %) is added into W1 Buffer and Wash Buffer when first open. Repeat the extraction procedure with a new sample.
	The volume or the percentage of ethanol is not correct before adding into W1 Buffer and Wash Buffer	Make sure that the correct volumes of ethanol (96- 100 %) is added into W1 Buffer and Wash Buffer when first use. Repeat the extraction procedure with a new sample.
	Elution of genomic DNA is not efficient	
	pH of water (ddH <sub>2</sub> O) for elution is acidic	Make sure the pH of ddH <sub>2</sub> O is between 7.5- 9.0.
		Use Elution Buffer (provided) for elution.
	Elution Buffer or ddH <sub>2</sub> O is not completely absorbed by column membrane	After Elution Buffer or ddH <sub>2</sub> O is added, stand the PGDE Column for 5 min before centrifugation.
<b>Column is clogged</b>		
	Sample is too viscous	Reduce the sample volume.
<b>Degradation of eluted DNA</b>		
	Sample is old	Always use fresh or well-stored sample for genomic DNA extraction.
	Buffer for gel electrophoresis contaminated with DNase	Use fresh running buffer for gel electrophoresis.