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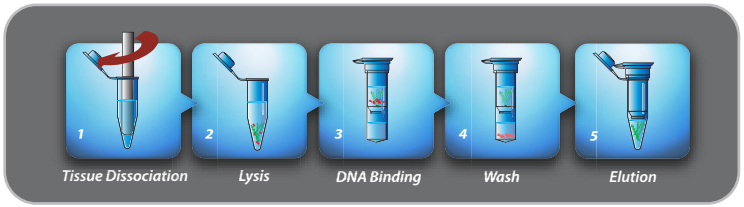
Real Genomics™

Store at room temperature 15°C~25°C



HiYield™ Genomic DNA Mini Kit (Plant) Protocol Book

Extraction of high quality DNA from plant tissues within 30 minutes

Cat. No. **YGP100**

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Real Genomics™

HiYield™ Genomic DNA Mini Kit (Plant)

Store at room temperature 15°C~25°C

Cat.No. **YGP100**
100 mini preps / kit
GP1 Buffer: 50 ml
GPX1 Buffer: 50 ml
GP2 Buffer: 15 ml
GP3 Buffer (concentrated): 30 ml *
W1 Buffer: 45 ml
Wash Buffer (concentrated) : 25 ml **
Elution Buffer: 30 ml
RNase A (10 mg/ml): 550 µl
GP Column : 100 pcs
Lysate Filter Column: 100 pcs
2 ml Collection Tube: 200 pcs

Sample: Up to 100 mg of fresh plant tissue, up to 25 mg of dry plant tissue
Yield: 3-5 µg (100 mg Arabidopsis thaliana leaf), 20-25 µg (100 mg Nicotiana tabacum leaf)
Format: Spin column (centrifuge or vacuum)
Operation Time: Within 30 minutes
Elution Volume: 30-200 µl

Mix following buffers prior to the initial use:
* Add isopropanol (see the bottle label for volume) to the GP3 Buffer immediately prior to initial use .
** Add absolute ethanol (see the bottle label for volume) to the Wash Buffer prior to initial use .

Description

HiYield™ Genomic DNA Mini Kit (Plant) provides a fast and simple method to isolate total DNA(genomic DNA, mitochondrial and chloroplast) from a wide variety of plant species and tissue types. Sample are distrusted by grinding in liquid nitrogen and lysed by lysis buffer incubation. Since different plant species contain different metabolites, such as polysaccharides, polyphenolics and proteins, two different lysis buffers(GP1 Buffer & GPX1 Buffer) are provided for various samples. The protocol does not require DNA phenol extraction and alcohol precipitation. The entire procedure can be completed in 30 minutes.

Features

Duo lysis buffer system for different metabolites.
Rapid extraction of ready-to-use DNA from plant tissues within 30 minutes.
Complete removal of all contaminants for sensitive downstream applications.

Applications

Purified DNA are ready for direct use in PCR, Southern Blotting, Real-Time PCR, AFLP, RFLP, PADP.

Quality Control

The quality of HiYield™ Genomic DNA Mini Kit (Plant) is tested on a lot-to-lot basis by isolation of genomic DNA from 50 mg of young leaf. Purified DNA is quantified with a spectrophotometer and the yield of genomic DNA is more than 20 µg with A_{260}/A_{280} ratio 1.7 - 1.9. The purified DNA is checked by electrophoresis.

Storage

HiYield™ Genomic DNA Mini Kit (Plant) should be stored dry at room temperature (15–25°C) for up to 2 years without showing any reduction in performance and quality.

Reference: Vogelstein, B., and Gillespie, D. (1979) Proc. Natl. Acad. Sci. USA 76, 615.
Note: For research use only. This kit contains irritant agent. During operation, always wear a lab coat, disposable gloves and protective goggles.

Important Notes

1. Add isopropanol (see the bottle label for volume) to the GP3 Buffer immediately prior to initial use .
2. Add absolute ethanol (see the bottle label for volume) to the Wash Buffer prior to initial use .
3. Additional requirements:
   - 96% ~ 100% Absolute Ethanol.
   - Sterile, DNase-free pipette tips and 1.5 ml microcentrifuge tubes.
   - Isopropanol.
   - ddH_2O.

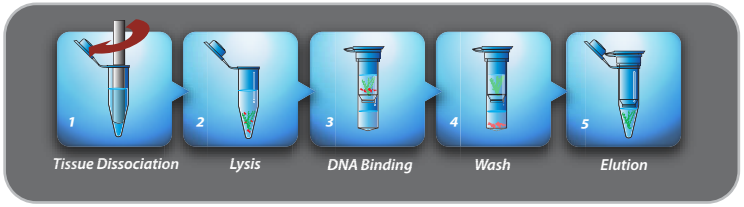
Protocol

Important Technical Information:

Since different plant species contain different metabolites, such as polysaccharides, polyphenolics and proteins, two different lysis buffers (GP1 Buffer & GPX1 Buffer) are provided for various samples.

GP1: GP1 Buffer is suitable for most common plant species. This buffer system ensures purified DNA with high quality and high yield.
GPX1: An alternative buffer, GPX1, is also included with this kit. The detergent present in this buffer is more effective in dispersing plant samples with large amounts of polysaccharide.

For the majority of plant species, both buffers will give similar results. The researcher may try one buffer system first or both in parallel.



Tissue Dissociation

1. Cut off 50 mg (up to 100 mg) of fresh or frozen plant tissue or 10 mg (up to 25 mg) of dried sample.
2. Grind the sample under liquid nitrogen to a fine powder with pestle and mortar.
3. Transfer it into a 1.5ml microcentrifuge tube.

NOTE: Some plant samples can be ground sufficiently in the absence of liquid nitrogen.

Lysis

4. Add 400 µl of GP1 Buffer (or GPX1 Buffer) and 5 µl of RNase A (10 mg/ml) into the sample tube and mix by vortexing.
   Mix GP1 Buffer or GPX1 Buffer and RNase A immediately prior to use. Do not mix them before use.
5. Incubate at 60°C for 10 minutes. During incubation, invert the tube every 5 minutes. At the same time, preheat required Elution Buffer (200 µl per sample) at 60°C.
6. Add 100 µl of GP2 Buffer and mix by vortexing.
7. Incubate on ice for 3 minutes. Place a Lysate Filter Column in a 2 ml Collection Tube and apply the mixture from previous step to the Lysate Filter Column.

Lysis

8. Centrifuge for 1 minutes at 1,000 x g then discard the Lysate Filter Column.
9. Carefully transfer the supernatant from the 2 ml Collection Tube to a new 1.5ml microcentrifuge tube.

DNA Binding

10. Add 1.5 volumes of GP3 Buffer (isopropanol added) to the cleared lysate and mix immediately by vortexing for 5 seconds. For example, add 750 µl of GP3 Buffer to 500 µl of lysate. If precipitate appears, break it up as much as possible with a pipette.
11. Place a GP Column in a new 2 ml Collection Tube.
12. Apply 700 µl of the mixture (including any precipitate) from previous step to the GP Column.
13. Centrifuge at full speed (14,000~16,000 rpm) for 2 minutes.
14. Discard flow-through in the 2 ml Collection Tube and apply remaining mixture to GP Column.
15. Centrifuge at full speed (14,000~16,000 rpm) for 2 minutes.
16. Discard flow-through then place the GP Column back in the 2 ml Collection Tube.

Wash

17. Add 400 µl of W1 Buffer into the GP Column.
18. Centrifuge at full speed (14,000~16,000 rpm) for 30 seconds.
19. Discard the flow-through and place the GP Column back in the 2 ml Collection Tube.
20. Add 600 µl of Wash Buffer (ethanol added) into the GP Column.
21. Centrifuge at full speed (14,000~16,000 rpm) for 30 seconds.
22. Discard the flow-through and place the GP Column back in the 2 ml Collection Tube.
23. Centrifuge again at full speed (14,000~16,000 rpm) for 3 minutes to dry the column matrix.

Optional Step: Residue Pigment Removal

If a few pigment remains on the column matrix, perform this optional step.
a. Following the Wash Buffer addition, add 400 µl of absolute ethanol in the GP Column.
b. Centrifuge at full speed (14,000~16,000 rpm) for 30 seconds.
c. Discard the flow-through and place the GP Column back in the 2 ml Collection Tube.
d. Centrifuge again at full speed (14,000~16,000 rpm) for 3 minutes to dry the column matrix.

DNA Elution

24. Transfer dried GP Column into a clean 1.5 ml microcentrifuge tube.
25. Add 100 µl of preheated Elution Buffer or TE into the center of the column matrix.
    Standard elution volume is 100 µl. If less sample to be used, reduce the elution volume (30~50 µl) to increase DNA concentration. If higher DNA yield is required, repeat the DNA Elution step to increase DNA recovery and the total elution volume is about 200 µl.
26. Stand for 3-5 minutes until Elution Buffer or TE absorbed by the matrix.
27. Centrifuge at full speed (14,000~16,000 rpm) for 30 seconds to elute purified DNA.

HiYield™ Genomic DNA Mini Kit (Plant)

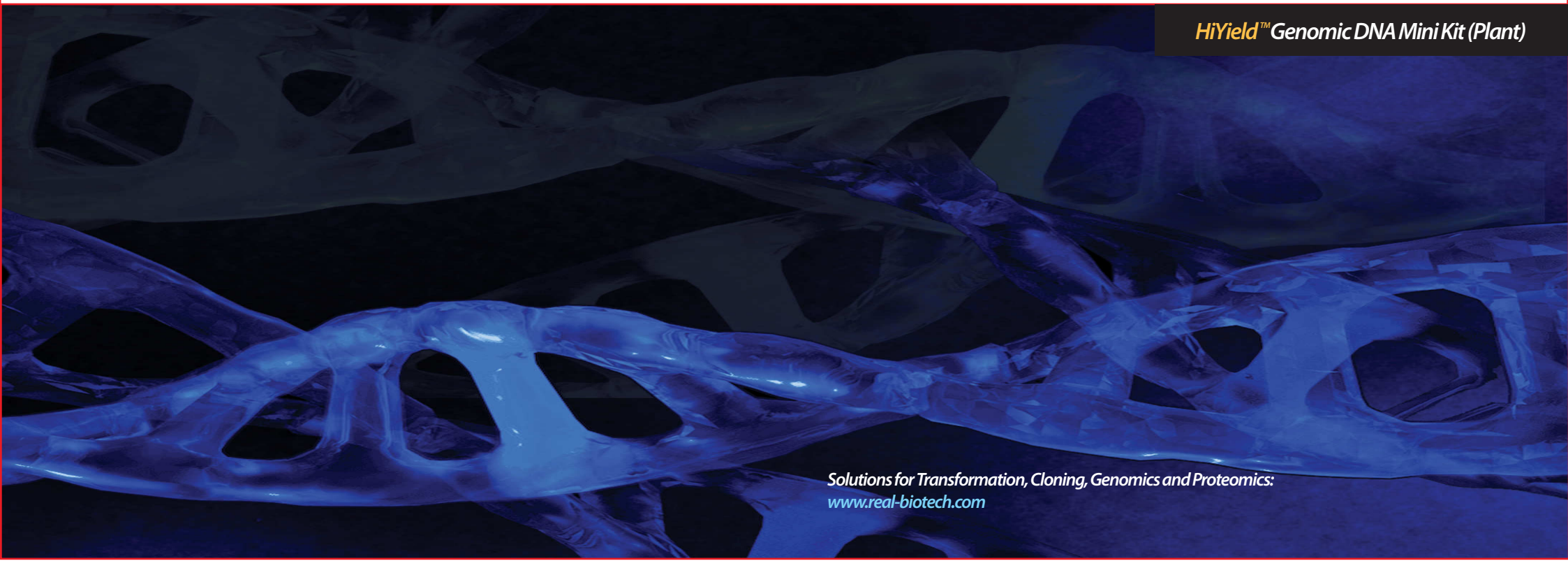
Ordering Information

		Cat. No.	Size	Items	Contents
Genomic DNA	Blood& Cultured Cells	YGB100	S	HiYield™ Genomic DNA Mini Kit (Blood / Bacteria/ Cultured Cells)	100 preps/kit,(RBC Lysis, GB, GT, Wash, W1, Elution) Buffer...etc.
		YGB300	S	HiYield™ Genomic DNA Mini Kit (Blood / Bacteria/ Cultured Cells)	300 preps/kit,(RBC Lysis, GB, GT, Wash, W1, Elution) Buffer...etc.
		YGBI25	M	HiYield™ Genomic DNA Midi Kit (Fresh Blood / Cultured Cells)	25 preps/kit,(RBC Lysis, GB, Wash, W1, Elution) Buffer...etc.
		YGDI25	M	HiYield™ Genomic DNA Midi Kit (Frozen Blood / Cultured Cells)	25 preps/kit,(GB, Wash, W1, Elution) Buffer, Proteinase K...etc.
		YGBM10	L	HiYield™ Genomic DNA Maxi Kit (Fresh Blood / Cultured Cells)	10 preps/kit,(RBC Lysis, GB, Wash, W1, Elution) Buffer...etc.
		YGDM10	L	HiYield™ Genomic DNA Maxi Kit (Frozen Blood / Cultured Cells)	10 preps/kit,(GB, Wash, W1, Elution) Buffer, Proteinase K...etc.
	Tissue	YGT50	S	HiYield™ Genomic DNA Mini Kit (Tissue)	50 preps/kit,(GT, GB, Wash, W1, Elution) Buffer, Proteinase K...etc.
		YGT100	S	HiYield™ Genomic DNA Mini Kit (Tissue)	100 preps/kit,(GT, GB, Wash, W1, Elution) Buffer, Proteinase K...etc.
		YGT300	S	HiYield™ Genomic DNA Mini Kit (Tissue)	300 preps/kit,(GT, GB, Wash, W1, Elution) Buffer, Proteinase K...etc.
	Plant	YGP100	S	HiYield™ Genomic DNA Mini Kit (Plant)	100 preps/kit,(GP1, GPX1, GP2, GP3, Wash, W1, Elution) Buffer, RNase A...etc.
		YGPI25	M	HiYield™ Genomic DNA Midi Kit (Plant)	25 preps/kit,(GP1, GPX1, GP2, GP3, Wash, W1, Elution) Buffer, RNase A...etc.
		YGPM10	L	HiYield™ Genomic DNA Maxi Kit (Plant)	10 preps/kit,(GP1, GPX1, GP2, GP3, Wash, W1, Elution) Buffer, RNase A...etc.
	96-Well	YGP96B-2	-	HiYield™ 96-Well Plant Genomic DNA Extraction Kit	2 preps/kit, (GP1, GPX1, GP2, GP3, Wash, W1, Elution) Buffer, RNase A,Genomic DNA Binding Plate,350ul Collection Plate...etc.
		YGP96B-4	-	HiYield™ 96-Well Plant Genomic DNA Extraction Kit	4 preps/kit, (GP1, GPX1, GP2, GP3, Wash, W1, Elution) Buffer, RNase A,Genomic DNA Binding Plate,350ul Collection Plate...etc.
		YGP96B-10	-	HiYield™ 96-Well Plant Genomic DNA Extraction Kit	10 preps/kit, (GP1, GPX1, GP2, GP3, Wash, W1, Elution) Buffer, RNase A,Genomic DNA Binding Plate,350ul Collection Plate...etc.
	Other	YVM96	-	Vacuum Manifold	Maximum Operating Vacuum: 28 in. Hg

Notes

HiYield™ Genomic DNA Mini Kit (Plant)

Notes



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