TIANGEN®

TGuide Smart Hi-Plant DNA Kit

(Prefilled single sample cartridge)

For extraction of genomic DNA from plant tissues, especially plant materials rich in polysaccharides, polyphenols or starch.

TECHNICAL MANUAL

Cat. no. GDP617-DE

Note: To use the TGuide Smart Hi-Plant DNA Kit, you must have the TGuide Smart Hi-Plant DNA Kit (program no. DP617) installed on the TGuide S16/S32 Pro/S96 Dex Nucleic Acid Extractor.

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Table Contents

TGuide Smart Hi-Plant DNA Kit

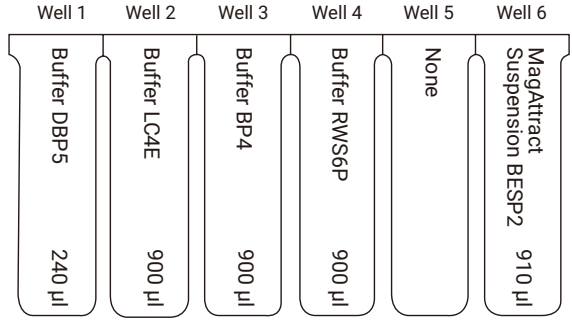
Cat. no. GDP617-DE

Kit Contents

	Contents	GDP617-DE (48 preps)
GDP617-DH	Buffer GPH	45 ml
	NKY tablets	3 tablets
	Proteinase K	1 ml
	Buffer ST	3×1 ml
	RNaseA (100 mg/ml)	250 μl
	Buffer TB	15 ml
	Hi-Plant DNA Reagents	48 pcs
OSE-TGA-S36	TGuide Smart Tip Comb	12 pcs

Note: GDP617-DH and OSE-TGA-S36 are shipped and stored separately.

Hi-Plant DNA Reagents composition



Storage

Store at room temperature (15-30℃) in a dry environment. If precipitates form in any buffer, incubate the solution in a 37℃ water bath for 10 minutes to fully dissolve precipitates.

Introduction

This kit adopts specially functionalized magnetic beads paired with delicately formulated buffer systems to isolate and purify high-quality genomic DNA from a wide range of plant tissues, especially plant materials rich in polysaccharides, polyphenols or starch, as well as macrofungal specimens. Optimized polyphenol removal tablets efficiently eliminate complex secondary metabolites contained in plant samples.

Compatible with fully automated nucleic acid extractors including TGuide S16 / S32 Pro / S96 Dex, the uniquely coated magnetic beads exhibit strong affinity for nucleic acids under defined buffer conditions; upon condition modulation, captured nucleic acids are eluted from beads to realize rapid nucleic acid separation and purification. Custom magnetic combs capture, transfer and release magnetic beads to complete nucleic acid translocation, enabling high-throughput automated extraction.

The whole extraction workflow is safe and user-friendly. The purified genomic DNA is characterized by high purity, large fragment size, and consistent quality. DNA obtained using this kit is suitable for a variety of downstream applications, including restriction enzyme digestion, PCR amplification, library preparation, Southern blot analysis, microarray assays, high-throughput sequencing, and third-generation sequencing.

Features

Broad Compatibility: Suitable for genomic DNA extraction from a wide range of polyphenol- and polysaccharide-rich plant tissues and fungal samples.

Safe and Non-toxic: No toxic organic reagents such as phenol or chloroform required.

Simple and Rapid: Ultra-pure genomic DNA can be obtained within 1 hour.

High Purity: Purified DNA can be directly used for microarray analysis and high-throughput sequencing.

Notes <u>Please read these notes carefully before using this kit.</u>

1. Add NKY tablets into Buffer GPH before use: add one NKY tablet into 21 ml Buffer GPH for extraction of 24 samples. Select one of the following NKY tablets application protocols as appropriate for your experiment:

   1) For small batch of samples: Crush the tablet into pieces (mung bean-sized pieces); add approximately 40 mg of NYK tablet piece per sample, followed by Buffer GPH and vortex for 2 min before subsequent extraction.

   2) For large batch of samples: Dissolve one tablet in 21 ml Buffer GPH and vortex for 2 min to form a white opaque suspension (stable for 4 weeks at ambient temperature). Mix the solution thoroughly before each use. Minor unevenness

after dispensing into centrifuge tubes does not compromise experimental performance.

2. Avoid repeated freeze-thaw cycles of samples, which will result in shortened DNA fragments and reduced DNA yield.

Recommended Input Amounts for Various Sample Types

Sample Type	Recommended Input Amount	Example Sample Library
Common Leaves	60-100 mg	Rice, Maize, Wheat, Soybean, Phoenix tree, Reed, Sweet potato
Polysaccharide- & Polyphenol-Rich Leaves	30-60 mg	Strawberry, Willow, Eucalyptus, Smoketree, Acer truncatum, Pomelo, Maple, Chinese rose, Chrysanthemum, Hosta, Lonicera maackii, Mulberry leaf, Hawthorn, White poplar, Toona, Bodhi tree, Weigela
High-Moisture Tissues	100-300 mg	Chinese cabbage, Wild peach, Muskmelon
Flower Petals	60-150 mg	Rose, Chrysanthemum, Wild rose, Yellow iris, Yellow iris pollen, Pine pollen
Roots & Stems	100-200 mg	Salvia miltiorrhiza root, Curcuma root, Ginseng, Maize stalk, Reed stalk
Tubers	50-100 mg	Sweet potato, American ginseng
Seeds	30-60 mg	Peanut, Bean sprout, Ginseng, Barley, Maize
Traditional Chinese Medicinal Materials	30-60 mg	Curcuma, Salvia miltiorrhiza, Polygonatum, Fritillary bulb
Fungal Samples	30-60 mg	Hypsizygus marmoreus, White beech mushroom, Shiitake, Enoki mushroom, Dictyophora, Aspergillus niger

Protocol

1. Preparation of Hi-Plant DNA Reagents

1.1 Remove the single sample reagent cartridge from the kit, invert several times to resuspend the magnetic beads. Remove the outer packaging bag and lightly flick the strip to ensure that the reagents and magnetic beads are collected at the bottom. Carefully peel off the sealing film before use, avoid shaking to prevent liquid splashing.

1.2 Add appropriate volume (60-100 µl) of elution Buffer TB to well 5 before start.

2. Sample Preparation

2.1 Sample Grinding

a. Liquid nitrogen grinding: According to the recommended input amount for different sample types, take the plant tissue and grind it thoroughly in liquid nitrogen.

b. Homogenizer processing: Snap-freeze samples in liquid nitrogen, then transfer it to the steel bead grinding tubes (TIANGEN, Cat. no. OSE-TH-B04). Homogenize with the TGrinder H24 Tissue Homogenizer (TIANGEN, Cat. no. OSE-TH-01): oscillate at a speed of 6.5 m/s for 60 sec, with a 15-second interval, for a total of 4 cycles. For high-throughput grinding, the TGrinder LH96 can be used in combination with a 96-well grinding tube adapter (TIANGEN, Cat. no. OSE-TH-A05); For large-volume samples, various stainless steel jars (OSE-TH-A08 to 12) are available. Set oscillation frequency at 60 Hz and grind for 2 min with steel beads.

Note: When using homogenizers from other manufacturers, optimize grinding parameters in accordance with the instrument's official protocol.

2.2 Add 800 µl Buffer GPH **(confirm NKY tablets have been added beforehand)**, 50 µl Buffer ST and 20 µl Proteinase K to the sample. Vortex thoroughly (TGyrate Master Votex, Cat. no. OSE-VX-02 with accessory OSE-VX-M131 is recommended). Incubate at 60℃ for 10 min (TGrade Dry Bath Incubator, OSE-DB series are recommended). Invert the tube several times during incubation for sufficient sample mixing.

Note:

1) After NKY tablets are added into Buffer GPH and vortexed for 2 min, a white insoluble suspension forms. Mix thoroughly before each use. Slight uneven distribution when aliquoting into centrifuge tubes will not affect the experimental results.

2) Buffer GPH, Buffer ST and Proteinase K can be premixed proportionally right before use. Do not store the mixed solution for extended periods.

2.3 Centrifuge at 12,000 rpm (~13,400×g) for 3 min.

Note: Pipette only 600 µl supernatant for downstream experiments to avoid aspirating bottom impurities, discard residual liquid.

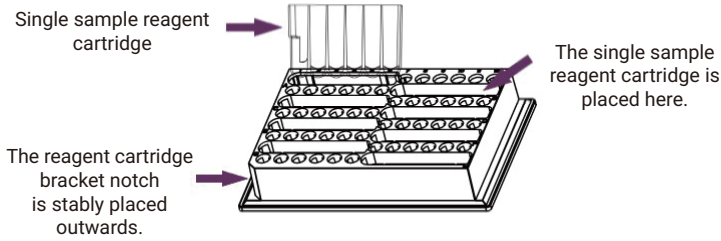
3. Operating Steps for TGuide S16/S32 Pro/S96 Dex Nucleic Acid Extractor

3.1 Transfer 600 µl of preprocessed supernatant into Well 1 of the single sample reagent cartridge, and add 5 µl RNaseA (100 mg/ml) into Well 2.

Note: RNaseA must be added into Well 2 immediately before instrument run to preserve enzymatic activity; prepare and add on-demand only.

3.2 Place the single sample reagent cartridge onto the dedicated single sample reagent cartridge holder.

3.3 Mount the single sample reagent cartridge holder onto the base platform inside TGuide S16/S32 Pro/S96 Dex instrument; insert tip combs into corresponding comb rack slots and fully lock the buckles in place.



3.4 Enter the program of the TGuide S16/ S32 Pro/S96 Dex, select the matching preset protocol file and click the Run button to start the experiment.

The detailed experimental procedures are shown in the table below:

Table 1: Hi-Plant DNA Extraction Program for TGuide S16 Automated Nucleic Acid Purification System

Step	Hole site	Step name	Mix time (min)	Mix speed	Dry time (min)	Volume (μl)	Temp. (°C)	Segments	Every time (sec)	Magnetization time(sec)	Cycle	Magnet speed (mm/s)
1	6	Transfer Beads	0	8	--	900	--	5	5	0	2	2.5
2	2	Retain Beads	1	8	--	900	--	0	0	0	0	2.5
3	1	Bind	5	8	--	900	--	0	0	0	0	2.5
4	2	Transfer Beads	0	8	--	900	--	5	5	0	2	2.5
5	1	Bind	5	8	--	900	--	5	10	0	2	2.5
6	2	Wash 1	0.5	8	--	100	--	1	0	0	1	2.5
7	2	Wash 2	6	8	--	900	--	5	5	0	2	2.5
8	3	Wash 3	0.5	8	--	100	--	1	0	0	1	2.5
9	3	Wash 4	2.5	8	--	900	--	5	5	0	2	2.5
10	4	Wash 5	0.5	8	--	100	--	1	0	0	1	2.5
11	4	Wash 6	2	8	--	900	--	5	5	0	2	2.5
12	6	Wash 7	0.5	8	--	100	--	1	0	0	1	2.5
13	6	Wash 8	2	8	6	900	--	5	5	0	2	2.5
14	5	Elution	6	8	--	100	75	5	12	0	2	2.5
15	6	Discard	0.5	8	--	300	--	1	0	0	1	2.5

Table 2: Hi-Plant DNA Extraction Program for TGuide S32 Pro Automated Nucleic Acid Purification System

Step	Hole Site	Step Name	Waiting time (min)	Mixing time (min)	Magnetic suction time (min)	Mixing speed	Volume (μl)	Temper-ature (°C)	Adsorption mode
1	6	Transfer Beads	0	0.5	1	Fast	910	--	Cycle
2	2	Retain Beads	0	0.5	0	Fast	900	--	---
3	1	Lysis	0	5	0	Fast	900	--	---
4	2	Transfer Beads	0	0	1	Fast	900	--	Cycle
5	1	Mix	5	5	1	Fast	900	--	Cycle
6	2	Wash 1	0	0.5	1	Fast	100	--	---
7	2	Wash 2	0	6	1	Fast	900	--	Cycle
8	3	Wash 3	0	0.5	1	Fast	100	--	---
9	3	Wash 4	0	2.5	1	Fast	900	--	Cycle
10	4	Wash 5	0	0.5	1	Fast	100	--	---
11	4	Wash 6	0	2	1	Fast	900	--	Cycle
12	6	Wash 7	0	0.5	1	Fast	100	--	---
13	6	Wash 8	0	2	1	Fast	900	--	Cycle
14	5	Elution	6	6	2	Fast	100	75	Cycle
15	6	Discard	0	0.5	0	Fast	910	--	--

Table 3: Hi-Plant DNA Extraction Program for TGuide S96 Dex Automated Nucleic Acid Purification System

Step	Hole Step	Step Name	Waiting time before mixing (mm:ss)	Mixing Speed	Mixing time (mm:ss)	Mixing mode	Liquid volume (μl)	Magnetic suction time (mm:ss)	Number of magnetic aspira-tions	Heating temper-ature (°C)	Pause after comple-tion
1	6	Transfer Beads	0	Fast	0:30	Normal Mode	910	0:30	2	-	-
2	2	Retain Beads	0	Fast	0:30	Normal Mode	900	0:00	0	-	-
3	1	Mix	0	Fast	5:00	Normal Mode	900	0:00	0	-	-
4	2	Transfer Beads	0	Fast	0:30	Normal Mode	900	0:30	2	-	-
5	1	Bind	0	Fast	5:00	Normal Mode	900	0:30	2	-	-
6	2	Wash 1	0	Fast	0:30	Normal Mode	100	0:30	0	-	-
7	2	Wash 2	0	Fast	6:00	Normal Mode	900	0:30	2	-	-
8	3	Wash 3	0	Fast	0:30	Normal Mode	100	0:30	0	-	-
9	3	Wash 4	0	Fast	2:30	Normal Mode	900	0:30	2	-	-
10	4	Wash 5	0	Fast	0:30	Normal Mode	100	0:30	0	-	-
11	4	Wash 6	0	Fast	2:00	Normal Mode	900	0:30	2	-	-
12	6	Wash 7	0	Fast	0:30	Normal Mode	100	0:30	0	-	-
13	6	Wash 8	0	Fast	2:00	Normal Mode	900	0:30	2	-	-
14	5	Elution	6:00	Fast	6:00	Normal Mode	100	0:30	3	-	-
15	6	Discard	0	Fast	0:30	Normal Mode	910	0	0	-	-

3.5 After completion of automated extraction program, aspirate purified DNA from Well 5 of the single sample reagent cartridge and store the DNA under appropriate conditions.

Appendix

Related Products

1. Instrument and Accessories

Product name	Packing Size	Cat.No
TGuide S16 Nucleic Acid Extractor	1 set	OSE-S16-AM
TGuide Smart Magnetic Tip Comb	200 pieces/box	OSE-TGA-S03
TGuide Single Sample Tank Bracket	5 pieces/box	OSE-TGA-S32

2. TGuide Smart Reagents Kits

Product name	Cartridge	Cat.No
TGuide Smart Viral DNA/RNA Kit	48 preps	GDP604-DE
TGuide Smart Universal DNA Kit	48 preps	GDP605-DE
TGuide Smart Envir-DNA Kit	48 preps	GDP613-DE
TGuide Smart Hi-Plant DNA Kit	48 preps	GDP617-DE
TGuide Smart DNA Purification Kit	48 preps	GDP642-DE
TGuide Smart Micro Cell Total RNA Kit	48 preps	GDP670-DE
TGuide Smart Universal RNA Kit	48 preps	GDP671-DE
TGuide Smart Hi-Plant RNA Kit	48 preps	GDP672-DE
TGuide Smart Processed Food DNA Kit	48 preps	GDP680-DE
TGuide Smart Viral DNA/RNA Kit	96 preps	GDP604-E
TGuide Smart Universal DNA Kit	96 preps	GDP605-E
TGuide Smart Envir-DNA Kit	96 preps	GDP613-E
TGuide Smart Hi-Plant DNA Kit	96 preps	GDP617-E
TGuide Smart DNA Purification Kit	96 preps	GDP642-E
TGuide Smart Micro Cell Total RNA Kit	96 preps	GDP670-E
TGuide Smart Universal RNA Kit	96 preps	GDP671-E
TGuide Smart Hi-Plant RNA Kit	96 preps	GDP672-E
TGuide Smart Processed Food DNA Kit	96 preps	GDP680-E