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# MGIEasy

Plant gDNA Extraction Set

**Instructions for Use** 

Version: 3.0

Research Use Only

Wuhan MGI Tech Co., Ltd.

#### About the instructions for use

This instructions for use is applicable to MGIEasy Plant gDNA Extraction Set. The version of the instructions for use is 3.0 and the set version is 1.0.

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# **Revision history**

Version	Date	Description
3.0	September 12, 2024	<ul> <li>Updated some reagent component specifications.</li> <li>Updated sample amount requirements.</li> <li>Updated procedures of extracting the nucleic acids manually.</li> <li>Updated reagent and sample preparation procedures of automated nucleic acids extraction.</li> <li>Updated parameter table of automated nucleic acids extraction on NE384RS.</li> </ul>
2.0	November 17, 2023	Updated the operation part
1.0	June 27, 2023	Initial release

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Introduction Instructions for Use

# **Chapter 1 Introduction**

#### 1.1 Product name

MGIEasy Plant gDNA Extraction Set

#### 1.2 Specifications

Set name	Model	Component	Cat. No.	Specification
MGIEasy Plant gDNA Extraction Set	PDT-96	MGIEasy Plant gDNA Extraction Kit	940-001321-00	96 RXN/Set
Cat. No.: 940-001323-00		RNase A	940-001304-00	
MGIEasy Plant gDNA Extraction Set	PDT-384	MGIEasy Plant gDNA Extraction Kit	940-001322-00	384 RXN/Set
Cat. No.: 940-001324-00		RNase A	940-001303-00	

#### 1.3 Intended use

This set is used to extract high-quality and high-purity gDNA from leaves and seeds of plants rich in polysaccharides and polyphenols.

#### 1.4 Working principle

By using this product, salt ions with high concentration lyse and release DNA from plants. The released DNA is then captured by magnetic beads and washed by a specific wash buffer to remove proteins, salt and other impurities. After being dried, the DNA in magnetic beads is eluted by the elution buffer and high-purity DNA is obtained.

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## 1.5 Main components

Table 1 MGIEasy Plant gDNA Extraction Set (PDT-96) Cat. No.: 940-001323-00

Name	Component	Specification	Storage condition	Validity period	Transportation condition
	Buffer PL	68 mL/tube×1			
	Buffer PB	34.0 mL/tube×1			
	Buffer WB I	51.0 mL/tube×1			
MGIEasy Plant gDNA Extraction Kit	Buffer WB II	29.0 mL/tube×1	2 ℃ to 30 ℃	18 months	2 °C to 30 °C
Cat. No.: 940-001321-00	Buffer TE	15 mL/tube×1			
	Magnetic Beads T	2 mL/tube×1			
	Proteinase K	2 mL/tube×1			
RNase A Cat. No.: 940-001304-00	RNase A	1 mL/tube×1	2 °C to 8 °C		2 ℃ to 8 ℃

Table 2 MGIEasy Plant gDNA Extraction Set (PDT-384) Cat. No.: 940-001324-00

Name	Component	Specification	Storage condition	Validity period	Transportation condition
	Buffer PL	269 mL/tube×1	2 °C to 30 °C	18 months	2 ℃ to 30 ℃
	Buffer PB	135.0 mL/tube×1			
	Buffer WB I	202.0 mL/tube×1			
MGIEasy Plant gDNA Extraction Kit	Buffer WB II	116.0 mL/tube×1			
Cat. No.: 940-001322-00	Buffer TE	60 mL/tube×1			
	Magnetic Beads T	8 mL/tube×1			
	Proteinase K	8 mL/tube×1			
RNase A Cat. No.: 940-001303-00	RNase A	4 mL/tube×1	2 °C to 8 °C		2 °C to 8 °C

Applicable device Instructions for Use

# Chapter 2 Applicable device

- MGISP-960RS High-throughput Automated Sample Preparation System
- MGISP-NE384RS Automated Nucleic Acid Extractor

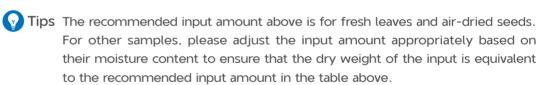
# Chapter 3 Sample requirements

#### 3.1 Applicable sample

This product is applicable to leaves and seeds of plants rich in polysaccharides and polyphenols.

#### 3.2 Sample amount requirements

	Common plant leaves	Polysaccharide-rich and polyphenol-rich plant leaves	Plant seeds
Sample amount	10 mg-100 mg	< 30 mg	10 mg-50 mg



# **Chapter 4 Operation**

#### 4.1 Preparing materials

Prepare the following materials:

Table 3 User-supplied materials

Туре	Item	Description
	Mini centrifuge	With a speed no less than 13000 rpm
	Vortex mixer	None
Equipment	Thermomixer compact	It can be replaced by a water bath
Equipment	1.5 mL magnetic rack	None
	Pipette	1 mL/200 μL/20 μL
	Grinding mill	None
Reagent	Absolute ethanol	Analytically pure
	Pipette tips	None
Consumable	Centrifuge tube	2 mL/1.5 mL
		• 3 mm
	Grinding media	• Zirconia
		RNase-free

#### 4.2 Extracting the nucleic acids

- Tips To realize high concentration and purity, please select fresh and young plants, and plump seeds, and fully grind them before extraction.
  - You can extract the nucleic acids manually or on automation devices. For automated nucleic acid extraction, ensure that you prepare applicable consumables.

#### 4.2.1 Extracting the nucleic acids manually

Before extraction, add absolute ethanol into Buffer WB I and Buffer WB II according to the label.

Perform the following steps:

1. Freeze the fresh plant leaves with liquid nitrogen for 10 seconds, or store it at -80 °C for over 30 minutes. Plant seeds do not need to be flash-frozen; they can be ground directly.

- 2. Select one of the following methods to grind the samples:
  - Tips To avoid thawing the leaves, grind leaves quickly or in liquid nitrogen.
  - Place the plant seeds or leaves in a cooled mortar and use a pestle to grind them into powder. Transfer the powder into a new 1.5 mL or 2 mL centrifuge tube.
  - Place the plant seeds or leaves into a new 2 mL centrifuge tube. Add grinding media into the tube. Place the tube into a grinding mill to grind leaves into powder.
- 3. According to sample type, choose one of the following procedures:
  - Common plant leaves:

Immediately add 700  $\mu$ L of Buffer PL and 10  $\mu$ L of RNase A to the centrifuge tube. Mix thoroughly using a vortex mixer. Perform a warm bath in a thermomixer at 60 °C for 10 minutes with a rotation speed of 1000 rpm, or vortex for 10 seconds every 2 minutes.

Polysaccharide-rich and polyphenol-rich plant leaves:

Immediately add 500  $\mu$ L of Buffer PL and 10  $\mu$ L of RNase A to the centrifuge tube. Mix thoroughly using a vortex mixer. Perform a warm bath in a thermomixer at 60 °C for 25 minutes with a rotation speed of 1000 rpm, or vortex for 10 seconds every 2 minutes. After lysis, add 110  $\mu$ L of absolute ethanol to the centrifuge tube, mix thoroughly using a vortex mixer. Place on ice for 10 minutes, and vortex once during this period.

Plant seeds:

Immediately add the reagents to the centrifuge tube in the following order: first, 20  $\mu$ L of proteinase K, then 700  $\mu$ L of Buffer PL, and finally 10  $\mu$ L of RNase A. Mix thoroughly using a vortex mixer. Perform a warm bath in a thermomixer at 60 °C for 10 minutes with a rotation speed of 1000 rpm, or vortex for 10 seconds every 2 minutes.

- Tips Common plant leaves include leaves of tomato, tobacco, rice, wheat, corn, citrus, soybean, etc.
  - Polysaccharide-rich and polyphenol-rich plant leaves include leaves of cherry, peach, kiwi, strawberry, peanut, ginkgo, azalea, oak, pine needles, etc.
  - If the sample type is uncertain, it is recommended to follow the polysaccharide and polyphenol extraction procedure.

- 4. Place the tube into a centrifuge at 13000 rpm for 7 minutes.
- 5. Carefully transfer all the supernatant to a new 1.5 mL centrifuge tube, and avoid transferring any solid impurities.
- 6. Add Buffer PB with half the volume of the supernatant, 20 µL of Magnetic Beads T, and 20 µL of Proteinase K to the centrifuge tube (Proteinase K is only for leaf samples, it is not needed for seed samples). Place the centrifuge tube in a vortex mixer and mix thoroughly, then place it at room temperature for 6 minutes, and vortex for 10 seconds every 2 minutes.
- 7. Place the tube on the magnetic rack for 30 seconds. Invert the magnetic rack and collect the magnetic beads on the wall and at the bottom of the tube. When Magnetic Beads T is adsorbed completely on the tube wall, use a pipette to remove the supernatant.
- 8. Remove the centrifuge tube from the magnetic rack. Add 700 µL of Buffer WB I into the centrifuge tube. Vortex the tube for 30 seconds and place the tube on the magnetic rack for 30 seconds. Invert the magnetic rack and collect the magnetic beads on the wall and at the bottom of the tube. When Magnetic Beads T is adsorbed completely on the tube wall, use a pipette to remove the supernatant.
- 9. Remove the centrifuge tube from the magnetic rack. Add 700 µL of Buffer WB II into the centrifuge tube. Vortex the tube for 30 seconds and place the tube on the magnetic rack for 30 seconds. Invert the magnetic rack and collect the magnetic beads on the wall and at the bottom of the tube. When Magnetic Beads T is adsorbed completely on the tube wall, use a pipette to remove the supernatant.
- 10. Repeat step 9 once. Decap and dry the tube for 10 minutes during which remove the liquid continuously until no liquid remains on the tube. Ensure that the surface of the magnetic beads is non-reflective and no cracks appear on the beads.
- 11. Remove the centrifuge tube from the magnetic rack. Add 60  $\mu$ L to 150  $\mu$ L of Buffer TE into the tube. Vortex the tube to mix thoroughly and incubate at room temperature for 5 minutes during which vortex the tube every 2 minutes with 10 seconds for each time.
- 12. Place the tube on the magnetic rack for 2 minutes. When Magnetic Beads T is adsorbed completely on the tube wall, aspirate the supernatant and transfer it into a new sterile centrifuge tube. The supernatant is the extracted DNA.

# 4.2.2 Extracting the nucleic acids automatically on MGISP-960RS

#### 4.2.2.1 Preparing consumables

According to the following table, prepare consumables for a workflow of automated extraction on MGISP-960RS and place them at room temperature until use.

Name	Brand	Cat. No.	Number
250 μL automated filter tips	MGI	100000723	5
2.2 mL V-bottom 96-well deep-well plate	MGI	1000008088	4
1.3 mL U-bottom 96-well deep-well plate	MGI	1000004644	1
Hard-shell thin-wall 96-well skirted PCR plates	MGI	1000012059	1

#### 4.2.2.2 Preparing reagents

- 1. Before extraction, add absolute ethanol into Buffer WB I and Buffer WB II according to the label.
- 2. Take out 5 96-well deep-well plates and a PCR plate. Add sample and reagents according to the following table:

Reagent name	Adding volume	Plate
Mix	<ul> <li>Buffer PB: half the volume of the supernatant</li> <li>Magnetic Beads T: 20 µL</li> <li>Proteinase K: 20 µL (Proteinase K is only required for leaf samples; it is not needed for seed samples)</li> </ul>	2.2 mL V-bottom 96-well deep- well plate
Buffer WB I	700 µL/well	2.2 mL V-bottom 96-well deep- well plate
Buffer WB II	1400 µL/well	2.2 mL V-bottom 96-well deep- well plate
Buffer TE	100 µL/well	1.3 mL U-bottom 96-well deep- well plate

Reagent name	Adding volume	Plate
Plate for waste (empty)	/	2.2 mL V-bottom 96-well deep- well plate
DNA product (empty)	/	Hard-shell thin-wall 96-well skirted PCR plates



- Tips When aliquoting reagent, add the reagent to the bottom of the plate to prevent the reagent from adhering to the walls.
  - The prepared Mix is required to be used within 30 minutes. If you want to prepare the Mix in advance, add Proteinase K before adding the Mix to the plate to avoid inactivating Proteinase K caused by long-time preparation.
  - The reagents for the Mix plate can be added after the supernatant is removed. They can be added separately or mixed before adding.

#### 4.2.2.3 Preparing samples

For detailed operations about preparing samples, refer to steps 1 to 4 in Extracting the nucleic acids manually on page 4.

After sample preparation, carefully transfer all the supernatant to the deep-well plate that contains the Mix, and avoid transferring any solid impurities.

#### 4.2.2.4 Starting extraction

- 1. Switch to the position to power on the device.
- 2. Turn on the computer and the desktop appears. Double-click it or un the software.

- 3. Select **User** and **Real**. Enter the password.
- 4. Click **Login** to enter the main interface.
- 5. On the upper-right corner of the control software, click and select WDesigner. The home interface is displayed.
- 6. Ensure that the application file in the .wfex format has been prepared.
- 7. Click in the toolbar and find the file location in the pop-up window.
- 8. Select the file and click Open, fill in the Application and Project, and click Confirm to save the application file. Then this application file can be executed in the control software.

9. After the file is imported successfully, click



in the toolbar.

- 10. Click **Initialize** on the top of the interface to start initializing. You will be prompted after a successful initialization.
- 11. Click the menu button on the left of the interface, and select **Clean>Pre- clean>Start**.
- 12. Follow the on-screen instructions to complete operations and click **Continue**. The UV lamp and air filter start working.
  - CAUTION The ultraviolet radiation is harmful to the human body, so do not open the door after the pre-clean starts.
- 13. Import the application scripts according to MGISP-100&MGISP-960 Application Script Installation Instructions.
- 15. Click the drop-down list of **Solution** and select **JB-A09-140 MGIEasy Plant gDNA Extraction Set\_RV1.0\_SV2.0**. Click the drop-down list of **Script** and select **JB-A09-140 MGIEasy Plant gDNA Extraction Set\_RV1.0\_SV2.0.py**. Place samples, reagents and consumables according to the following figure:

Name	Position
DNA product (empty plate)	POS12
Buffer TE	POS13
Buffer WB I	POS14
Buffer WB II	POS15
Plate for waste	POS16
Mix	POS20

Name	Position
250 µL automated filter tips	POS1~5

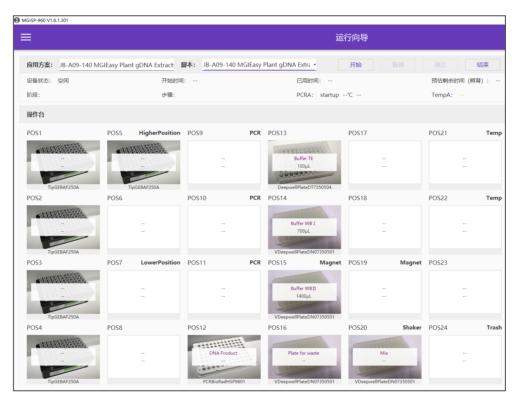


Figure 1 Plate position

- 16. Click **Start**. The extraction workflow starts. It takes roughly 1 hour.
  - During the workflow, click **Pause** to pause and click **Resume** to resume the workflow if required.
- 17. At the end of the workflow, remove the DNA product from POS12. If the product is not used immediately, seal and store it in a freezer at -80  $^{\circ}$ C .
- 18. Dispose of the used deep-well plates, PCR plates, and waste bag.

# 4.2.3 Extracting the nucleic acids automatically on MGISP-NE384RS

#### 4.2.3.1 Preparing consumables

According to the following table, prepare consumables for a workflow of automated extraction on MGISP-NE384RS and place them at room temperature until use:

Name	Brand	Cat. No.	Quantity
2.2 ml V bottom 06 well doon well plate	MGI	1000008088	5 (96 preps)
2.2 mL V-bottom 96-well deep-well plate		1000008088	20 (384 preps)
OF well tips samp	MGI	1000025661	1 (96 preps)
96-well tips comb		1000025001	4 (384 preps)

#### 4.2.3.2 Preparing reagents

- 1. Before extraction, add absolute ethanol into Buffer WB I and Buffer WB II according to the label.
- 2. Take out 5 96-well deep-well plates. Add sample and reagents according to the following table:

Reagent name	Volume of reagent to be added	Plate	
Mix	<ul> <li>Buffer PB: half the volume of the supernatant</li> <li>Magnetic Beads T: 20 µL</li> <li>Proteinase K: 20 µL (Proteinase K is only required for leaf samples; it is not needed for seed samples)</li> </ul>	2.2 mL V-bottom 96-well deep-well plate	
Buffer WB I	700 μL/well	2.2 mL V-bottom 96-well deep-well plate	
Buffer WB II	700 μL/well	2.2 mL V-bottom 96-well deep-well plate	
Buffer TE	100 μL/well	2.2 mL V-bottom 96-well deep-well plate	



Tips • When aliquoting reagent, add the reagent to the bottom of the plate to prevent the reagent from adhering to the walls.

- The prepared Mix is required to be used within 30 minutes. If you want to prepare the Mix in advance, add Proteinase K before adding the Mix to the plate to avoid inactivating Proteinase K caused by long-time preparation.
- The reagents for the Mix plate can be added after the supernatant is removed. They can be added separately or mixed before adding.

#### 4.2.3.3 Preparing samples

For detailed operations about preparing samples, refer to steps 1 to 4 in Extracting the nucleic acids manually on page 4.

After sample preparation, carefully transfer all the supernatant to the deep-well plate that contains the Mix, and avoid transferring any solid impurities.

#### 4.2.3.4 Starting extraction

- 1. Switch to the position to power on the device.
- 2. Turn on the computer and the desktop appears. Double-click the icon of MGISP-NE384RS to run the software.
- 3. Select User and Real, and enter the password. Click Login to enter the main interface.
- 4. Click **Initialize** on the top of the interface to start initializing. You will be prompted after a successful initialization.
- 5. Click **Process manage**> to import the script.
- 6. Click The Sworkflow. Click the drop-down list of Script and select MGIEasy Plant gDNA Extraction Set\_V2.0. Place samples, reagents and consumables according to the following table:

Reagent name	Position
Mix	POS1
Buffer WB I	POS2
Buffer WB II	POS3
Buffer WB II	POS4
Buffer TE	POS6

7. Click **Run**. The device starts extraction according to the following table. The whole workflow takes about 35 minutes.

During the workflow, click **Pause** to pause and click **Resume** to resume the workflow if required.

Heating settings are as follows:

Lysis temperature: 25 °C . Lysis heating ends at step 2.

Elution temperature: 25 °C. Elution heating starts at step 5.

Dayanastay	Step No.					
Parameter	Step 1	Step 2	Step 3	Step 4	Step 5	Step 6
Step Name	Lysis	Wash	Wash	Wash	Elution	Release
Pos	Pos 1	Pos 2	Pos 3	Pos 4	Pos 6	Pos 2
volume(µL)	940	700	700	700	120	700
Delay Time(s)	0	0	0	0	150	0
Mix	True	True	True	True	True	True
Mix Type	Normal	Normal	Normal	Normal	Normal	Normal
Mix Rate	HighMiddle	HighMiddle	HighMiddle	HighMiddle	HighMiddle	High
Mix Time(s)	240	180	120	120	300	3
Collect	True	True	True	True	True	False
Collect Mode	Cycle	Cycle	Cycle	Cycle	Cycle	/
Collect Cycle	3	3	2	2	45	/

- 8. After the program ends, transfer the 96-well tips comb with magnetic beads T to the medical waste bag.
- 9. Remove the 96-well plate from POS6 and transfer the DNA product to a new tube.

If the product is not used immediately, seal it and store it in a freezer at -80  $^{\circ}\text{C}$  .

10. Dispose of the used deep-well plates, PCR plates and waste bag.

## **Chapter 5 Warnings and precautions**

- This product is for research use only. Please read the instructions for use carefully before use.
- To realize high concentration and purity, please select fresh and young plants, and plump seeds, and fully grind them before extraction.
- The magnetic beads may be adhesive to the side or bottom of the tube during wash, which is a normal phenomenon and will not affect DNA extraction and downstream application.
- During extraction on MGISP-NE384RS, the magnetic beads may remain on the tube after elution. This will not affect the produce concentration and purity. You need to separate the beads again.
- Before experiment, be sure to be familiar with and master the operation methods and precautions of various devices to be used.
- Direct contact with skin and eyes should be avoided for all samples and reagents. Do not swallow. If accidental ingestion occurs, please get medical attention immediately. If skin exposure occurs, rinse with large amounts of water and get medical attention if irritation persists.
- All samples and wastes should be disposed of in accordance with relevant regulations.
- Do not use expired products.

# **Appendix 1 Manufacturer information**

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