Tinzyme Co., Limited

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Magbead Plant DNA Kit (96 Auto Plate)

Product Number: EK0306

Shipping and Storage

Store at 2-30℃.

Component

Component	EK0306
Buffer PLS	60mL
RNase A (10 mg/mL)	0.6mL
96 DW Auto Plate 1	1 Plate
96 DW Auto Plate 2	1 Plate
96 DW Auto Plate 3	1 Plate
96 DW Auto Plate 4	1 Plate
96 DW Auto Plate 5	1 Plate
96 DW Auto Plate 6	1 Plate
96 Tip Comb	1 Pc

Description

This kit provides a simple and efficient method for extracting plant DNA. After plant cells are physically broken, the DNA in the lysate binds to the surface of silicon-based coated magnetic beads in the presence of high salt. After rinsing, the DNA is washed in Buffer TB or deionized water. The extraction yield is closely related to the sample type and cell fragmentation effect, and the extracted DNA can be used for downstream experiments such as second-generation sequencing and PCR testing.

Self provided instruments and reagents

1. DNA Extractor.

Note

1. The sample should avoid repeated freezing and thawing, otherwise it may lead to a decrease in the amount of nucleic acid extracted or a smaller number of extracted nucleic acid fragments.
2. Before use, check if there is any leakage of reagents in the pre installed plate. Before using the pre installed plate, please mix it upside down and immediately remove or gently shake it to avoid liquid accumulation.

Protocol

1. Sample pretreatment (liquid nitrogen grinding or homogenizer grinding and homogenization);
2. Add Buffer PLS and RNase A; Incubate at 70℃, centrifuge to obtain supernatant;
3. Invert and mix pre packaged reagents, centrifuge or shake gently to avoid liquid accumulation;
4. Transfer all supernatant to 96 DW Auto Plate 1;
5. Insert the instrument and run the program;
6. Transfer nucleic acid from 96 DW Auto Plate 6 to obtain purified DNA.

Fragmentation of plant materials

1. Option 1

1.1. Weigh about 50-100mg of fresh plant sample powder that has been thoroughly ground with liquid nitrogen or about 30 mg of dried sample powder, quickly transfer it to a centrifuge tube pre filled with 600μL Buffer PLS and 5μL RNase A

(10 mg/mL), quickly invert and mix, centrifuge instantly, and place it in a constant temperature mixer at 70 ℃, 1600 rpm, for 10 minutes.

Note: Adding β - mercaptoethanol to Buffer PLS to extract polysaccharides and polyphenols from plants at a final concentration of 5% can improve the nucleic acid extraction efficiency.

2. **Option 2:**

2.1. Add 50-100mg of fresh plant material or 20 mg of dried plant material to a 2.0mL centrifuge tube;

2.2. After rapid freezing with liquid nitrogen, use a grinding pestle to thoroughly grind the plant materials into powder;

2.3. Add 600μL of Buffer PLS and 5μL of RNase A (10 mg/mL) to the centrifuge tube, quickly invert and mix, and place in a constant temperature mixer at 70℃, 1600 rpm, for 10 minutes.

Note: Adding β - mercaptoethanol to Buffer PLS to extract polysaccharides and polyphenols from plants at a final concentration of 5% can improve the nucleic acid extraction efficiency.

3. **Option 3**

3.1. Add 50-100mg of fresh plant material or 20mg of dried plant material to a 2.0mL centrifuge tube;

3.2. Add 600μL Buffer PLS, 5μL RNase A (10mg/mL), and 1 steel ball to the centrifuge tube, and immediately fix the centrifuge tube in the tissue disruptor for shaking and crushing;

3.3. After removing the centrifuge tube from the tissue disruptor, centrifuge it instantly and place it in a constant temperature mixer at 70℃, 1600 rpm, for 10 minutes.

Manual operation

1. After centrifugation at 12000 rpm (~13400 × g) for 4 minutes, transfer all the supernatant to 96 DW Auto Plate 1.
2. Place the deep hole plate in the corresponding position on the DNA Extractor, place the magnetic rod sleeve in 96 DW Auto Plate 4, and click to run the program
3. After about 45 minutes of program execution, transfer the elution product from 96 DW Auto Plate 6 to a 1.5mL centrifuge tube and store it at -20℃ for future use.

Result Analysis

The DNA concentration should be determined by measuring the absorbance at 260nm (A260) using a spectrophotometer. DNA has a significant absorption peak at 260nm, and the absorbance reading should be between 0.1 and 1.0. If it is not within this range, the sample needs to be diluted or concentrated. At 260nm, one unit of absorbance corresponds to 50μg of DNA per milliliter (A260=1 → 50μg/mL). The residual magnetic beads in the eluent may affect the A260 reading, but will not affect the performance of downstream DNA.

DNA sample concentration=50μg/mL × (A260-A320) × dilution factor

Total DNA amount=concentration x sample volume (mL)

The purity of DNA is determined by calculating the ratio of the corrected absorbance at 260nm to 280nm (subtracting the absorbance reading at 320nm to correct for the presence of magnetic particles), i.e. (A260-A320)/(A280-A320). The A260/A280 ratio of pure DNA is 1.7-1.9.