Product Information

Universal DNA Gel / PCR Purification Kit

Catalog Number: D2022

Product Size: 100T, 200T

Application Scope: DNA gel extraction, PCR or digestion purification

Contents:

Component	100T	200T
A : Binding Buffer	80 mL	160 mL
B : Wash Buffer Concentrate	25 mL	50 mL
C : Elution Buffer	6 mL	12 mL
D : Spin Column	100 pcs	200 pcs
E : Collection Tubes	100 pcs	200 pcs
Before use, please prepare wash buffer working solution (add 96-100% ethanol)		
Required EtOH	100 mL	200 mL

Storage

Store at room temperature. When stored as directed, product is stable for at least 12 months.

Description

US EVERBRIGHT Universal DNA Gel / PCR Purification Kit is a silica gel-based DNA extraction product. It can extract 65 bp-10 kb DNA fragments from TAE or TBE buffer. Each column can bind up to 20 μg of DNA. DNA Gel / PCR Purification Kit can effectively remove GelRed, GelGreen and other nucleic acid dyes in the gel, and the recovery rate can reach more than 80%. The purified DNA can be used in downstream experiments such as transformation, sequencing, PCR, and digestion et al..

Protocol

Preparing the experiment

● Before the first use, add an appropriate volume of 96-100% ethanol to Wash Buffer Concentrate. For example, add 25 mL Wash Buffer Concentrate to 100 mL ethanol, or 50 mL Wash Buffer Concentrate to 200 mL ethanol.

● Prepare 1.5 mL centrifuge tube

Method

Extracting DNA fragments from agarose gels

1. Cut an agar gel containing the target DNA fragment under UV, blot the gel surface liquid with a paper towel, and mince. Collect the gel in a 1.5 mL centrifuge tube (the weight of the extracted gel in a single tube must not exceed 300 mg).

2. Add 500 μL of Binding Buffer to the above 1.5 mL centrifuge tube.

3. Water bath at 55 ℃ for 5-10 minutes, until the gel is completely melted. Remove and mix to speed up melting during water bath.

4. After the gel is completely dissolved, check the color of the solution. If the solution is yellow, the pH is ideal. DNA can be bound to the silica gel column. If the solution is orange or red,

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10 uL 3M Acetic acid, sodium salt solution (pH 5.0) needs to be added to the solution to turn the solution yellow.

5. After the sample is returned to room temperature, transfer it to an spin column (the spin column is placed in a collection tube) and centrifuge at 11,000 × g for 30 seconds. Discard the filtrate. If there are many samples, centrifuge several times and discard the filtrate.

6. Return the spin column to collection tube, add 750 µL wash buffer (make sure that ethanol has been added), and centrifuge at 11,000 × g for 30 seconds. Discard the filtrate.

7. Return the spin column to collection tube, centrifuge at 11,000 × g for 2-3 minutes, and remove the remaining ethanol from the spin column (this step can choose the centrifuge at the maximum speed of the centrifuge, such as 18,000 × g. In order to completely remove the ethanol, the spin column can be left at room temperature for 5min to obtain better results).

8. Place spin column in a 1.5 mL new centrifuge tube and add 30-50 µL Elution Buffer or ddH_2O to the middle of the spin column membrane. Let it stand at room temperature for 1-5 minutes (If the DNA fragment is larger than 5 kb, please use Elution Buffer or ddH_2O heated to 60 ° C to improve extraction).

9. Centrifuge at maximum speed (eg 18,000 × g) for 1 min to collect purified DNA.

Purifying DNA from PCR product or digestion product

1. Estimate the volume of the PCR product or digestion product, add 5 times the volume of Binding buffer to it, and mix well.

Note: 1) For example, add 50 µL of PCR product to 250 µL of Binding Buffer.

2) The maximum purified volume does not exceed 100 µL. If the PCR product exceeds 100 µL, please use multiple spin column for purification.

2. Transfer the above solution to an spin column (put the spin column into a collection tube), leave it at room temperature for 1-2 minutes, and centrifuge at 11,000 × g for 30 seconds. Discard the filtrate.

3. Return spin column to collection tube, add 750 µL wash buffer (make sure that ethanol has been added), and centrifuge at 11,000 × g for 30 seconds. Discard the filtrate.

4. Return spin column to collection tube, centrifuge at 11,000 × g for 2-3 minutes, and remove the remaining ethanol from the spin column. The spin column can be left at room temperature for 5 min to obtain better results).

5. Place the spin column in a 1.5 mL new centrifuge tube, and add 30-50 µL Elution Buffer or ddH_2O to the middle of the spin column membrane. Let stand at room temperature for 1-5 min

Note: If the DNA fragment is larger than 5 kb, please heat the Elution Buffer or ddH_2O to 60 ℃ to improve the purified rate.

6. Centrifuge at maximum speed (such as 18,000 × g) for 1 min to collect purified DNA.

Notes

1. The binding buffer contains irritating compounds. Wear latex gloves and glasses during operation to avoid contamination of the skin, eyes and clothes. Mixing bleach and binding buffer can produce harmful by-products. Do not mix waste from the gel extract with bleach.

2. In step 1, cutting the gel into small pieces can greatly shorten the gel melting time (linear DNA is prone to hydrolysis under prolonged exposure to high temperature conditions), thereby improving extraction. Do not expose the gel containing DNA to UV light for a long time to reduce the damage to DNA caused by UV light.

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