



Ver. 260401

Gel Mini Purification Kit

Spin Column
(ODP208)

Kit Contents

Contents	50 Preps
Buffer GN	50mL
Buffer PW	15mL
Buffer EB	15mL
Spin Columns (CB3) & Collection Tubes	50 Nos

Storage

Gel Mini Purification Kit can be stored dry at room temperature (15-25°C) for up to 12 months without showing any reduction in performance and quality. For longer storage, the kit can be stored at 2-8°C.

(Note: Check buffers for precipitate before use and dissolve at 50°C for 10 minutes if necessary.)

Introduction

Gel Mini Purification Kit is designed to extract and purify DNA from any agarose gel in either TAE or TBE buffer without phenol extraction or ethanol precipitation. The kit combines the convenience of spin-column technology with the selective binding properties of a uniquely designed silica membrane. Special buffers are optimized for efficient recovery of DNA and removal of contaminants. The binding capacity of spin column CB3 is 10µg DNA per column. Purified DNA by the kit can be directly used in applications such as restriction enzyme digestion, PCR amplification, sequencing, library screening, ligation and transformation and so on.

Important Notes

1. Add 60mL ethanol (96-100%) to Buffer PW before use.
2. All centrifugation steps are carried out in conventional tabletop microcentrifuge at room temperature.
3. If the purification efficiency is low, check solution pH after agarose gel dissolved completely. If pH ≥ 7.0 , add 10µL of 3M sodium acetate (pH 5.0) to the gel solution until the solution pH adjusted between 5-7.
4. The recovery efficiency is related to starting DNA quantity and elution volume. The less starting quantity or elution volume, the less recovery efficiency.

Protocol

Note: Prepare an aliquot of Buffer EB and pre-warm at 60°C for 2 minutes before adding to the spin column (step 9).

1. Excise the DNA fragment from the agarose gel and transfer it to a clean 1.5mL microcentrifuge tube. Weight the gel slice and add **THREE** volumes of Buffer GN. If the weight of gel is 50mg, add 150µL Buffer GN.

**Note: For $\geq 2\%$ agarose gels, add SIX volumes of Buffer GN
Cut as close to the DNA as possible to minimize the gel volume.**

2. Incubate at 50°C for 10 minutes and mix the tube by inversion every few minutes to facilitate the melting process.

Note: After the gel dissolve, ensure that the colour of lysate is yellow, as it indicates the pH required for binding to spin column. If it is not yellow, add 10µL of 3M sodium acetate, pH 5.0 (not provided in the kit) to it and mix to change the colour to yellow.

3. After the mixture is cooled to room temperature, add ONE volume of isopropanol and mix by inverting the tube 2 times.
4. Place a spin column in a 2mL collection tube. Add solution from Step 3 into spin column. Centrifuge 7000 rpm for 1 minutes.
5. To get high yield DNA add flow-through solution into spin column and repeat the step 4.
6. Discard the flow- through, add 500µL Buffer GN to the spin column. Centrifuge at 7000 rpm for 1 minutes. Discard the flow-through and re-place the spin column.
7. Add 750µL Buffer PW to the spin column. Centrifuge at 13,000 rpm for 30 seconds. Discard the flow-through and re-place the spin column.
8. Centrifuge at 13,000 rpm for 3 minutes to remove residues. Discard the collection tube and place the spin columns to a new 1.5mL microcentrifuge tube (not provided).
9. Add 30-50µL Buffer EB. Incubate the mixture for 1 minute at room temperature. Centrifuge at 13,000 rpm for 2 minutes and discard the spin column.
10. Alternatively, for increased DNA concentration, add the solution gained from step 9 to the center of membrane again, let the columns stand for 1 minute, and then centrifuge.

Note: The volume of Buffer EB must be at least 30µL, or it may affect recovery efficiency. What's more, the pH value of eluted buffer will have some influence in eluting, we suggest chose Buffer EB or distilled water (pH 7.0-8.5) to elute DNA. For long-term storage of DNA, eluting in Buffer EB and storing at -20°C is recommended, since DNA stored in water is subject to acid hydrolysis.