

PCR Mini Purification Kit

Spin Column
(ODP203)

Kit Contents

Contents	50 Preps
Buffer PN	30mL
Buffer PW	15mL
Buffer EB	15mL
Spin Columns (CB3) & Collection Tubes	50

Storage

PCR 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 37°C for 10 minutes if necessary.)

Introduction

PCR Mini Purification Kit is based on silica-membrane technology. The kit is specially designed for purification of DNA fragments from various reaction solutions (PCR reactions, enzymatic reaction, etc) by removing contaminants of protein, other organic compound, salts and primers, etc. The recovery yield is more than 80% for 100bp-10kb DNA fragments. The binding capacity of column CB3 is 5µg DNA per column. DNA purified by the kit can be directly used in applications such as restriction enzyme digestion, PCR amplification, sequencing, library screening, ligation and transformation.

Important Notes

1. Add 60mL ethanol (96-100%) to Buffer PW before use.
2. All centrifugation steps are carried out at 13,000 rpm in conventional tabletop microcentrifuge at room temperature.
3. The recovery efficiency is related to starting DNA quantity and elution volume. The less starting quantity or elution volume, the less recovery efficiency.

4. The kit has no selectivity for the DNA fragment purification. If you want to purify specific DNA fragment selectively and remove some DNA fragment, use Gel Mini Purification Kit (ODP208).

Protocol

Prepare an aliquot of Buffer EB and pre-warm at 60°C for 2 minutes before adding to the spin column (step 7)

1. Mix **one volume** of PCR sample with **three volumes** of Buffer PN and **two volumes** of isopropanol (for example, **20µL** PCR sample + **60µL** Buffer PN + **40µL** Isopropanol)

Note: 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 3M Sodium acetate, pH 5.0 (not provided in the kit) to it and mix to change the colour to yellow.

2. Place a spin column CB3 in a 2mL collection tube. Add solution from Step 1 into spin column and incubate at room temperature for 3 minutes. Centrifuge at 7,000 rpm for 1 minute. Discard the flow through and re-place the spin column to 2mL collection tube.
3. To get high yield DNA add flow-through solution into spin column and repeat the step 2.
4. Discard the flow-through, add 500µL Buffer PN and centrifuge at 7,000 rpm for 1 minutes.
5. 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.
6. 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).
7. Add 30-50µL Buffer EB. Incubate the mixture for 1 minutes at room temperature. Centrifuge at 13,000 rpm for 2 minutes and discard the spin column.
8. Alternatively, for increased DNA concentration, add the solution gained from step 7 to the center of membrane again, let the column stand for 1 minute, and then centrifuge.

Note: The volume of Buffer EB must be more than 30μL, or it may affect recovery efficiency. What's more, the pH value of eluted buffer will have some influence in eluting, we suggest 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.