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LiPure™ DNA Clean-up Kit

Cat. #: M0036 Size: 200 rxn

Introduction

LiPure™ DNA Clean-up Kit utilizes a new silica membrane technology and reagent formulation to recover 100 bp to 10 kb of DNA fragments from PCR product or enzymatic reactions (such as restriction enzyme digestion, ligation and probe labelling). This kit uses a unique spin column to bind DNA, followed by washing and eluting steps. Each column can bind up to 10 µg of DNA, while maximumly removing impurities such as primers, oligonucleotides, and enzymes.

The purified DNA has high purity and concentration, good integrity, and the recovery rate is high. The purified DNA can be directly used for molecular biology experiments such as sequencing, ligation and transformation, labeling, and in vitro transcription.

Package Information

Components	M0036
Buffer PB	120 ml
Buffer PS	60 ml
Buffer PW (concentrated)	25 ml
Buffer EB	30 ml
Spin Columns DM with Collection Tubes	200

Not included in the kit: 100% ethanol

Storage

Store at Room temperature (15-30°C)

Preparation before the experiment and precautions

1. All components can be stored in a stable, dry, room temperature (15-30 °C) environment for 1 year, and at 2-8°C for longer storage. When the solution is stored at low temperature, it should be left at room temperature for a period of time to return to room temperature before use.

2. The kit can recover all the DNA fragments in the solution. If you need to selectively recover specific fragments and remove other different size fragments, please use our company's gel recovery kit.

3. 100% ethanol must be added to the Buffer PW before the first use according to the instructions on the bottle label.

4. Please check If crystallization or precipitation occurs in the Buffer PG before use. If yes, place it in a 37°C water bath for 3-5 minutes until it becomes clear.

5. The recovery efficiency is related to the initial amount of DNA and the elution volume. The smaller the initial amount, the smaller the elution volume and the lower the recovery efficiency.

6. All centrifuge steps can be done at room temperature.

Protocol

1. Estimate the volume of the reaction, add 5 volumes of Buffer PB and mix thoroughly.

Note: 1) if the volume of the reaction is 50 ul, add 250 ul Buffer PB.

2) Check the PH value of the solution after adding Buffer PB. If the PH >7.5, add 10-30 ul 3M Sodium acetate to adjust it to 5-7.

2. Column equilibration: Add 200 µl Buffer PS to the column (Spin Columns DM with collection tube). Centrifuge at 13,000 rpm for 1 minute. Discard the waste from the collection tube and put the column back to the collection tube.

3. Add the solution obtained in step 1 to a spin column with the collection tube and leave it at room temperature for 1 minute. Centrifuge at 13,000 rpm for 30-60 s, and discard the waste, then put the column back to the collection tube.

Note: The maximum volume of the column is 750 µl. If the sample volume is greater than 750 µl, it can be added by multiple times.

4. Add 500 µl Buffer PW (check if 100% ethanol has been added before use) to the column. Centrifuge for 30-60 s at 13,000 rpm. Discard the waste and return the column to the collection tube.

Note: If the recovered DNA is used for salt-sensitive experiments (such as blunt-end ligation or direct sequencing), it is recommended to add Buffer PW and let it stand for 2-5 minutes before centrifugation.

5. Centrifuge at 13,000 rpm for 1 minute and discard the waste from the collection tube. Leave the column at room temperature for a few more minutes to dry it completely.

Note: The purpose of this step is to remove the residual ethanol in the column, and the residual of ethanol will affect the downstream enzymatic reactions (enzyme digestion, PCR, etc.).

6. Place the column in a new 1.5 ml centrifuge tube (selfprepared) and add 30-50 µl of Buffer EB in the middle of the membrane and allow it to stand at room temperature for 1 minute. Centrifuge at 13,000 rpm for 1 minute and collect the DNA solution. Store the DNA at -20°C.

Note: 1) The pH of the elution buffer has a great influence on the elution efficiency. If water is used as elution buffer, the pH should be between 7.0 and 8.5 (use NaOH to adjust).

2) In order to increase the amount of DNA recovered, the solution obtained can be added back to column, and leave it at room temperature for 2 minutes, then centrifuge at 13,000 rpm for 1 minute.

3) The elution volume should not be less than 30 µl. Too little volume will affect recovery efficiency.

4) When DNA fragments are larger than 10 kb, Buffer EB should be preheated in a 50°C water bath to increase recovery efficiency.

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