

## Universal DNA purification and recovery kit

Item No.: HDP209 100 times  
HRP209M 200 times

Kit composition		100Time	200Time
Binding fluid GMB	Room temperature	100ml	200 ml
Rinsing liquid PE	Room temperature	50ml	100ml
Rinsing liquid WB	Room temperature	25 ml	50 ml
		Add the specified amount of ethanol according to the instructions before the first use	
Elution buffer EB	Room	15 ml	15 ml
Adsorption column	Room	100个	200个
Collection tube	Room	100个	200个

### Storage matters:

1. All solutions should be clear. If the solution may form a precipitate when the ambient temperature is low, it should not be used directly at this time. It can be heated in a 37°C water bath for a few minutes to restore clarity. It should be restored to room temperature before use.
2. Storage at low temperature (4°C or -20°C) will cause solution precipitation and affect the use effect, so transportation and storage are carried out at room temperature (15°C-25°C).
3. Avoid long-term exposure of the reagents to the air to cause volatilization, oxidation, and pH changes. The lid should be closed in time after each solution is used.

### Product description:

In the presence of highly isolated salts, the DNA fragments are selectively adsorbed on the silicon matrix membrane in the spin column, and then through a series of rapid rinsing-centrifugation steps, the rinsing fluid will remove the primers, nucleotides, proteins and enzymes. After the impurities are removed, the elution buffer with low salt and high pH will elute the pure DNA from the silicon matrix membrane.

### Features:

1. The silicon matrix membrane in the centrifugal adsorption column all adopts imported special adsorption membrane, the adsorption amount difference between the column and the column is very small, and the repeatability is good.
2. The use of high-quality sol solution does not contain the sodium iodide and perchlorate of traditional sol solution, and does not inhibit downstream reactions such as digestion, ligation and cloning after recovery.

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3. The sol solution is prepared by adding phenol red to make it a yellow color, which is convenient for observing the sol effect and monitoring the change of pH value to achieve the best combination effect and greatly improve the recovery efficiency.

4. The improved sol-liquid formula greatly improves the buffering capacity and stability, even if the sample changes greatly, the pH can be buffered within the optimal binding range.

5. Fast and convenient, no need to use toxic phenol, chloroform and other reagents, and no need for ethanol precipitation.

#### Precautions

a. All centrifugation steps are completed at room temperature, and the centrifuge speed needs to reach 13,000 rpm.

b. The sol liquid contains irritating compounds. Wear latex gloves during operation to avoid contaminating the skin, eyes and clothes. If it contaminates the skin or eyes, rinse immediately with plenty of water or saline.

c. The recovered and purified DNA fragments are generally between 100bp and 40kb, and the recovery efficiency of too long and too short fragments is rapidly reduced.

d. The amount of recovered DNA is related to the amount of starting DNA, elution volume, and DNA fragment size. Generally 1-15 $\mu$ g, 100bp-5kb DNA fragments, the recovery rate can be as high as 85%.

e. When cutting gum and recycling, UV light observation has a damaging effect on DNA fragments. Low-energy long-wave ultraviolet light should be used as much as possible, and the processing time under ultraviolet light should be shortened as much as possible.

f. The eluent EB does not contain the chelating agent EDTA, which does not affect downstream digestion, connection and other reactions. Water can also be used for elution, but it should be ensured that the pH is greater than 7.5. If the pH is too low, the elution efficiency will be affected. For elution with water, DNA fragments should be stored at -20°C. If DNA fragments need to be stored for a long time, they can be eluted with TE buffer (10mM Tris-HCl, 1mM EDTA, pH 8.0), but EDTA may affect the downstream digestion reaction, so it can be diluted appropriately when using.

#### Steps

Reminder: Please add the specified amount of absolute ethanol to the rinsing solution WB before using it for the first time. After adding, please tick to mark that ethanol has been added.

1. Under a long-wave UV lamp, use a clean blade to cut the DNA bands to be recovered, and try to cut off the DNA-free gel.

2. Put the excised gel containing DNA bands into a 1.5ml centrifuge tube and weigh it.

Weigh an empty 1.5ml centrifuge tube first, then put the gel block and weigh it again, subtract the weight of the two times to get the weight of the gel

3. Add 3 times the volume of binding solution GMB.

If the gel weighs 100 mg, its volume can be regarded as 100  $\mu$ l, then add 300  $\mu$ l of the sol solution.

If the gel concentration is greater than 2%, 6 times the volume of the sol solution should be added.

4. Place in a 56°C water bath for 10 minutes (or until the glue is completely dissolved). Vortex every 2-3 minutes to help accelerate dissolution.

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5. Optional: add 150µl of isopropanol for every 100mg of the initial gel weight, shake and mix well.

Sometimes adding isopropanol can improve the recovery rate, do not centrifuge after adding. When recovering fragments larger than 4Kb, do not add isopropanol, and sometimes it may reduce the recovery efficiency.

6. Add the solution obtained in the previous step to the adsorption column EC (the adsorption column is placed in the collection tube), leave it at room temperature for 1 minute, centrifuge at 12,000 rpm for 30-60 seconds, and discard the waste liquid in the collection tube.

If the total volume exceeds 750µl, the solution can be added to the same adsorption column EC twice.

7. Add 600µl rinsing solution WB (please check if absolute ethanol has been added first!), centrifuge at 12,000rpm for 30 seconds, discard the waste solution

8. Add 600µl of rinsing solution WB, centrifuge at 12,000 rpm for 30 seconds, and discard the waste solution.

9. Put the adsorption column EC back into the empty collection tube and centrifuge at 12,000 rpm for 2 minutes to remove the rinsing liquid as much as possible to avoid residual ethanol in the rinsing liquid from inhibiting downstream reactions.

10. Take out the adsorption column EC, put it into a clean centrifuge tube, add 50µl of elution buffer EB to the middle of the adsorption membrane (the elution buffer is heated in a 65-70°C water bath beforehand for better effect), and place it at room temperature Centrifuge for 1 minute at 12,000 rpm for 2 minutes. If a larger amount of DNA is needed, add the resulting solution to the adsorption column and centrifuge for 1 minute.

The larger the elution volume, the higher the elution efficiency. If a higher DNA concentration is required, the elution volume can be appropriately reduced, but the minimum volume should not be less than 25µl. If the volume is too small, the DNA elution efficiency will be reduced and the yield will be reduced.

(2) The following are the DNA purification steps such as PCR products or digested fragments.

1. Add 500µl binding solution GMB to every 100µl PCR amplification system or enzyme digestion system, and mix well. (If the initial system is less than 100µl, please adjust to 100µl with double distilled water in advance).

2. Add the solution obtained in the previous step to the adsorption column EC (the adsorption column is placed in the collection tube), leave it at room temperature for one minute, centrifuge at 12,000 rpm for 30-60 seconds, and discard the waste liquid in the collection tube.

**Note: If the total volume exceeds 750µl, add the solution to the same adsorption column EC twice. Optional step: add 500µl of protein-removing solution PE, centrifuge at 12,000rpm for 30-60 seconds, and discard the waste solution.**

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