

Micro universal DNA purification and recovery kit

Item No.: HDP210 100 times
HDP210M 200 times

Kit composition	save	100Time	200time
Balance solution	Room Temperature	5ml	20ml
Sol / binding liquid GMB	Room Temperature	100ml	200ml
Rinsing liquid WB	Room Temperature	25ml	50ml
		Add the specified amount of ethanol according to the instructions before the first use	
Rinsing liquid PE	Room	50ml	100ml
Micro adsorption column	Room	100A	200A
Elution buffer EB	Room	15ml	15ml
Collection tube (2ml)	Room	100A	200A

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 recover its clarification. 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:

This product is a special kit for the concentration and recovery of trace DNA. It is suitable for agarose gel DNA recovery of trace DNA, PCR reaction product purification and recovery, enzyme digestion product DNA fragment purification and recovery, probe labeling and purification and recovery, DNA sample concentration, etc. 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 special washer-free spin column design ensures that there is no liquid residue and contamination after centrifugation. Ensure the high purity of recovered DNA.

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2. The special micro spin column design can elute at a minimum of 10ul, ensuring a high concentration of recovered DNA.
3. 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.
4. The unique sol solution/binding solution formula unifies the two functions of sol and binding. Therefore, a kit can be used in agarose DNA recovery, PCR product cleaning and purification, enzyme digestion product purification and recovery, etc., saving the need The cost of purchasing multiple kits.
5. The sol liquid/binding liquid is adjusted to yellow, 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.
6. 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

Precautions:

1. The recovered and purified DNA fragments are generally between 100bp and 40kb, but within this range, the recovery efficiency decreases rapidly as the fragment length is too long or too short.
2. The amount of recovered DNA is related to the amount of starting DNA, elution volume, and DNA fragment size. For DNA fragments of 100bp-5kb, the clean recovery rate of PCR products can be as high as 85%-95%. If the fragment is too long, too short, and the recovery rate is low, you can choose a small volume elution (minimum not less than 5ul) to increase the concentration.
3. 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.
4. The eluent EB does not contain the chelating agent EDTA, and does not affect downstream reactions such as digestion and connection. 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.

About the use of balancing fluid

1. Introduction: Nucleic acid adsorption silica gel membrane column will react with the charge/dust in the air during long-term placement and affect its nucleic acid binding ability. After the silica gel column is pretreated with the balance solution, the hydrophobic groups of the silica gel membrane in the column can be greatly reduced, and the binding capacity of nucleic acid can be improved. Thereby improving the recovery efficiency or yield of the silica gel column. The balance solution is a strong alkaline solution. If you accidentally touch it, please wash it with a large amount of tap water. Close the cap tightly after use to avoid contact with air. Store at room temperature. Precipitation may be formed during storage. Please heat to 37°C to make the precipitation disappear completely.

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2. How to use: Take a new silica gel membrane adsorption column and install it in the collection tube, and draw 50 μ l of equilibration buffer into the column. Centrifuge at 13000 rpm for 1 minute, discard the waste liquid in the collection tube, and put the adsorption column back into the collection tube. At this time, the pretreatment of the column with the equilibration solution is complete. Follow the subsequent steps.

Steps:

Tip: Please add the specified amount of absolute ethanol to the rinse solution WB before using it for the first time. After adding it, please tick to mark that ethanol has been added to avoid multiple additions!

1. Agarose gel DNA recovery: Under long-wave ultraviolet light, use a clean blade to cut off the DNA bands to be recovered, and try to cut off the DNA-free gel. The smaller the gel volume, the better.

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 sol/binding solution GMB.

If the gel weighs 100 mg, its volume can be regarded as 100 μ l, then add 300 μ 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.

5. Optional, generally not required: add 150 μ l of isopropanol per 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.

Equilibrium solution pretreatment adsorption column: pretreatment of silica gel membrane adsorption column with equilibrium solution is a necessary step. For specific methods, please refer to the previous section "About the use of equilibrium solution"

6. Add the solution obtained in the previous step to the micro spin column (the adsorption column is placed in the collection tube), leave it at room temperature for 1 minute, centrifuge at 12,000 rpm for 30 seconds, and discard the waste liquid in the collection tube. If the total volume exceeds 750 μ l, add the solution to the same micro spin column twice. After the filtered sol/binding solution is mixed with the strong alkaline balance solution remaining in the collection tube, the sol solution may change from yellow to orange or even purple. This is the normal color change of the phenol red PH indicator under alkaline conditions.

Optional step: add 500 μ l of protein-removing solution PE, centrifuge at 12,000 rpm for 30-60 seconds, and discard the waste solution.

7. Add 500 μ l of rinsing solution WB (please check if absolute ethanol has been added!), centrifuge at 12,000 rpm for 30 seconds, and discard the waste solution.

8. Add 500 μ l of rinsing solution WB, centrifuge at 12,000rpm for 2 minutes, carefully take out the spin-dried micro centrifuge column and put it into a clean centrifuge tube.

This step combines the rinsing and spin drying spin column into one. When removing the micro spin column, be careful not to let the bottom edge of the spin column touch the rinsing liquid waste

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in the collection tube. In order to avoid residual ethanol in the rinse solution inhibiting downstream reactions.

9. Carefully align the pipette tip with the micro spin column. Add (10 μ l-30 μ l) Elution Buffer EB (elution buffer) to the middle part of the adsorption membrane (do not add to the tube wall)

The elution buffer is heated in a 65-70°C water bath in advance for better results), placed at room temperature for 1 minute, and centrifuged at 12,000 rpm for 1 minute. The larger the elution volume, the higher the elution efficiency. If higher DNA concentration is required, the elution volume can be appropriately reduced, but the minimum volume should not be less than 10 μ l. If the volume is too small, it will reduce the DNA elution efficiency and reduce the yield.

DNA purification such as PCR products or digested fragments:

1. Add 100 μ l sol/binding solution GMB to every 20 μ l PCR amplification system or enzyme digestion system, and mix well.

Note: Processing sample volume: sol binding solution volume = 1:5

Equilibrium solution pretreatment adsorption column: Pretreatment of silica gel membrane adsorption column with equilibrium solution is a necessary step. For specific methods, please refer to the previous section "About the use of equilibrium solution"

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

3. From this step, it is exactly the same as the operation steps 7-9 of agarose gel DNA recovery. Please refer to the operation steps 7-9 of agarose gel DNA recovery.

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