

Version No. FDL1720

DNA/RNA Extraction Kit (Column)

(Spin Column)

Catalog No.: D901

Kit Content:

Product composition	D901 (50 preps)
Buffer BL	30 ml
Buffer PB	25 ml
Buffer DW2	12 ml
Buffer EB	15 ml
FineBind Min-Elute DNA Spin Columns	50pcs
2 ml Collection Tubes	50pcs

Storage:

The kit can be stored at room temperature (15-25°C) under dry conditions for 12 months. There may be precipitation in Buffer PB at low temperature. Redissolve it in 37 °C water bath and shake well before use.

This Kit Special for Scientific Research.

Product introduction:

This kit is based on centrifugal adsorption column technology and is suitable for the recovery of 50bp-30Kb DNA fragments from agarose gels at various concentrations. In addition, the kit is also suitable for the recovery and purification of DNA from PCR products, enzymatic reaction solutions, or from crude DNA (including genomic DNA) obtained by various methods. Buffer PB contains pH indicator and the solution is yellow, which is convenient to determine whether the pH value of the solution is suitable for binding with DNA adsorption column. The efficiency of DNA recovery can be up to 80%, and the purified DNA can be directly used for sequencing, ligation, digestion, PCR, labeling, etc.

Precautions:

1. The addition of Buffer BL can improve the adsorption capacity of the adsorption column, improve the homogeneity and stability of the adsorption column, and eliminate the influence of high temperature/humidity or other adverse environmental factors on the adsorption column. Check whether Buffer BL is turbid before use. If so, it can be heated in 37°C water bath for a few minutes to restore clarification.
2. The Buffer PB contains yellow pH indicator indicating $\text{pH} \leq 7.5$.

Operation Steps:

Before use, add anhydrous ethanol to Buffer DW2 in the volume specified on the bottle.

Recover DNA fragments from agarose gel

1. Column equilibration steps: add 500 μl Buffer BL to the adsorption column FineBind Min- Elute DNA Spin Columns (put the adsorption column into the collection tube), centrifuge at 12,000 rpm for 1 min, dump the waste liquid in the collection tube, and put the adsorption column back into the collection tube. (Use the column treated on the same day)
2. Cut a single target DNA band from the agarose gel (cut off the excess as much as possible), place it in a clean centrifuge tube and weigh it.
3. Add equal volume of Buffer PB to the gel block (add 100 μl Buffer PB if the gel is 0.1 g and its volume can be deemed as 100 μl), place in a 50°C water bath for about 10 min and turn the centrifuge tube upside down gently in this period to ensure that the gel block is fully dissolved.

(Cut the gel block into pieces in advance if it is too large).

Note: For recovery of fragments smaller than 150 bp, the volume of Buffer PB can be increased to 3 times to improve the recovery rate; it is best to reduce the solution temperature to room temperature before transferring to the column after the gel block has completely dissolved, because the adsorption column has a strong ability to bind DNA at room temperature. The solution shall be yellow after the gel is completely dissolved before the follow-up operation. If the solution is orange or purple after the gel is completely dissolved, adjust the solution color to yellow with 10 μ l 3M sodium acetate (pH 5.0) before proceeding. (Buffer PB contains pH indicator. When pH \leq 7.5, the solution is yellow, and at this time DNA can effectively bind to the membrane. When pH value is high, the solution turns to orange and purple, which needs to be adjusted.)

4. Add the solution from the last step to the adsorption column (put the adsorption column into the collection tube), centrifuge at 12,000 rpm for 30 sec, discard the waste liquid in the collection tube and put the adsorption column in the collection tube.

Note: The volume of the adsorption column is 700 μ l. If the sample volume is greater than 700 μ l, it can be added in different times.

5. Add 500 μ l Buffer DW2 (**add anhydrous ethanol before use**) to the adsorption column, centrifuge at 12,000 rpm for 30 sec, discard the waste liquid in the collection tube and put the adsorption column in the collection tube.
6. Repeat step 5.
7. Centrifuge at 12,000 rpm for 3 min.
8. Put the adsorption column in a clean centrifuge tube, dropwise add an appropriate amount of Buffer EB to the middle part of the adsorption film, place at room temperature for 1 min, centrifuge at 12,000 rpm for 1 min and collect the DNA solution.

Note: The volume of eluent shall not be less than 30 μ l. Too small volume will affect the efficiency of recovery. If the downstream experiments are sensitive to pH, the sterile water may be used for elution. The pH value of the eluent has a great influence on the elution efficiency. If water is used as eluent, its pH value shall be between 7.0 and 8.5 (NaOH can be used to adjust the pH value of water to this range). When the pH value is lower than 7.0, the elution efficiency is not high.

Recover DNA from PCR reaction solution or enzyme digestion reaction solution

1. Column equilibration steps: add 500 μ L Buffer BL to the adsorption column FineBind Min- Elute DNA Spin

Columns (put the adsorption column into the collection tube), centrifuge at 12,000 rpm for 1 min, dump the waste liquid in the collection tube, and put the adsorption column back into the collection tube. (Use the column treated on the same day)

2. Calculate the volume of the PCR reaction solution or the enzyme digestion reaction solution, add the equal volume of Buffer PB and mix fully (no need to remove paraffin oil or mineral oil).

Note: For recovery of fragments smaller than 150 bp, the volume of Buffer PB can be increased to 3 times to improve the recovery rate; the solution shall be yellow after mixing before the follow-up operation. If the solution is orange or purple, adjust the solution color to yellow with 10 μ l 3M sodium acetate (pH 5.0) before proceeding.

3. Add the solution from the last step to the adsorption column (put the adsorption column into the collection tube), place in the room temperature for 2 min, centrifuge at 12,000 rpm for 30 sec, discard the waste liquid in the collection tube and put the adsorption column in the collection tube.

Note: The volume of the adsorption column is 700 μ l. If the sample volume is greater than 700 μ l, it can be added in different times.

4. Add 500 μ l Buffer DW2 (**add anhydrous ethanol before use**) to the adsorption column, centrifuge at 12,000 rpm for 30 sec, discard the waste liquid in the collection tube and put the adsorption column in the collection tube.
5. Repeat step 4 once.
6. Centrifuge at 12,000 rpm for 3 min.
7. Put the adsorption column in a clean of centrifuge tube, dropwise add an appropriate amount of Buffer EB to the middle part of the adsorption film, place at room temperature for 1 min, centrifuge at 12,000 rpm for 1 min and collect the DNA solution.

Note: The volume of eluent shall not be less than 30 μ l. Too small volume will affect the efficiency of recovery. If the downstream experiments are sensitive to pH, the sterile water may be used for elution. The pH value of the eluent has a great influence on the elution efficiency. If water is used as eluent, its pH value shall be between 7.0 and 8.5 (NaOH can be used to adjust the pH value of water to this range). When the pH value is lower than 7.0, the elution efficiency is not high.



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