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Gel Extraction Kit

Product Number: DRK0102

Shipping and Storage

1. All solutions should be clear. If the ambient temperature is low and the solution may form a precipitate, it should not be used directly. It can be heated in a 37℃ water bath for a few minutes to restore clarity. It should be restored to room temperature before use.
2. Storing at low temperatures (4℃ or -20℃) can cause solution precipitation, affecting the effectiveness of use. Therefore, transportation and storage are carried out at room temperature.
3. To avoid the volatilization, oxidation, and pH changes of reagents exposed to air for a long time, the lid of each solution should be promptly closed after use.

Components

Component	Storage	DRK0101 50Preps	DRK0102 100 Preps	DRK0103 200Preps
Balance Buffer	RT	5mL	10mL	20mL
Buffer DD	RT	40mL	75mL	150mL
Buffer WB	RT	13mL	25mL	50mL
		Add the specified amount of ethanol according to the bottle label instructions before the first use		
Buffer EB	RT	10mL	15mL	15mL
Adsorption column EC	RT	50	100	200
Collection tube(2mL)	RT	50	100	200

This reagent kit can be stored at room temperature for 12 months without affecting its effectiveness.

Description

In the presence of highly sequenced salt, DNA fragments after dissolution of agarose gel are selectively adsorbed on the silicon matrix membrane in the centrifuge column, and then through a series of rapid rinsing centrifugation steps, the rinsing solution will remove primers, nucleotides, proteins, enzymes and other impurities, and finally the low salt, high pH elution buffer will elute pure DNA from the silicon matrix membrane.

Features

1. All the silicon matrix membranes in the centrifugal adsorption column are made of specially designed adsorption membranes, with minimal differences in adsorption capacity between columns and good repeatability. Overcoming the drawbacks of unstable membrane quality in domestic reagent kits.
2. High quality sol solution was used, without the traditional sodium iodide and perchlorate of sol solution, which does not inhibit downstream reactions such as post harvest enzyme digestion and ligation cloning.
3. The addition of phenol red to the sol solution resulted in a yellow color, making it easier to observe the sol effect and monitor changes in pH value to achieve the best binding effect, greatly improving the yield efficiency.
4. The improved sol-gel formula greatly enhances buffering capacity and stability, allowing for pH buffering within the optimal binding range even with significant sample changes, without the need for acetic acid to adjust pH.
5. Fast, convenient, and does not require the use of toxic reagents such as phenol and chloroform, nor does it require ethanol precipitation.

Note

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1. All centrifugation steps are completed at room temperature using a traditional desktop centrifuge with a speed of up to 12000 rpm.
2. The sol solution and equilibrium solution contain irritating compounds. Latex gloves should be worn during operation to avoid contact with the skin and eyes. If it comes into contact with the skin or eyes, immediately rinse with plenty of water or saline solution.
3. The collection efficiency of purified DNA fragments is generally between 70bp and 40kb, and the collection efficiency of long and short fragments rapidly decreases. This kit has outstanding leading collection efficiency even when collecting 70bp fragments.
4. The amount of DNA collected is related to the amount of starting DNA, elution volume, and DNA fragment size. Generally, DNA fragments of 1-15μg and 100 bp-5 kb can achieve a yield of up to 85%.
5. When cutting and collecting glue, 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.
6. The eluent EB does not contain chelating agent EDTA, which does not affect downstream enzyme digestion, ligation 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, it will affect the elution efficiency. Wash with water, DNA fragments should be stored at -20°C. If DNA fragments need to be stored for a long time, they can be washed with TE buffer (10mM Tris HCl, 1mM EDTA, pH 8.0), but EDTA may affect downstream enzyme digestion reactions and can be diluted appropriately when used.

Protocol

Note: Tip: Before the first use, please add the specified amount of anhydrous ethanol to the Buffer WB bottle according to the label instructions. After adding, please mark the added ethanol with a hook in a timely manner to avoid multiple additions!

1. Under the long wave ultraviolet light, use a clean blade to cut off the DNA strips to be collected, and try to cut off the gel without DNA, so as to get the smaller the gel volume, the better.
2. Cut the cut gel containing DNA strips into small pieces and put it into a 1.5mL centrifuge tube for weighing.
   Weigh an empty 1.5mL centrifuge tube first, then put in the gel block and weigh again. Subtract the two weights to get the weight of gel.
3. Add 3 times the volume of sol solution DD.
   If the gel weighs 100 mg and its volume can be regarded as 100μL, add 300μL of sol solution.
   If the gel concentration is greater than 2%, add 6 times the volume of sol solution.
4. Place in a 56°C water bath for 10 minutes (or until the gel is completely dissolved). Vortex oscillation every 2-3 minutes helps accelerate dissolution.
5. Optional, generally not required: add 150μL isopropanol every 100 mg of initial gel weight, shake and mix well.
   Sometimes adding isopropanol can improve yield, do not centrifuge after adding. When collecting fragments larger than 4Kb, not adding isopropanol may sometimes actually reduce the collection efficiency.
6. Column Balance: Add 100μL Balance Buffer to Adsorption Column EC, centrifuge at 12000 rpm for 1 minute, discard the filtrate, and set aside for later use.
   Balance Buffer can enhance the adsorption capacity of silica gel membrane for nucleic acids. Please use the adsorption column processed on the same day.
7. Add the solution obtained in the previous step to the Adsorption column EC (place the adsorption column in the collection tube), let it stand at room temperature for 1 minute, centrifuge at 12000 rpm for 1 minute, and discard the filtrate.
   If the total volume exceeds 750μL, the solution can be added twice to the same adsorption column EC.
   After mixing the filtered sol solution with the strong alkaline equilibrium solution remaining in the collection tube, the sol solution may change from yellow to orange red or even purple, which is a normal color change under alkaline conditions of phenol red pH indicator.

8. Add 600μL Buffer WB (please check if anhydrous ethanol has been added first!) Centrifuge at 12000 rpm for 30 seconds and discard the filtrate. Add 600μL Buffer WB and rinse again, discard the filtrate.
9. Empty the adsorption column into the collection tube, centrifuge at 12000 rpm for 2 minutes, and try to remove the Balance Buffer as much as possible to avoid residual ethanol in the Balance Buffer inhibiting downstream reactions.
10. Remove the adsorption column and place it in a clean centrifuge tube. Add 50μL of elution buffer EB (the elution buffer can be preheated in a water bath at 80℃-90℃ to increase yield) to the middle of the adsorption membrane. Let it stand at room temperature for 2 minutes, centrifuge at 12000 rpm for 1 minute, and discard the adsorption column.

    Recommendation: To increase the efficiency of DNA collection, the obtained solution can be re added to the centrifugal adsorption column, left at room temperature for 1 minute, and centrifuged at 12000 rpm for 1 minute. Washing twice can increase the concentration by about 10%.

    The larger the elution volume, the higher the elution efficiency. If a higher DNA concentration is required, the elution volume can be appropriately reduced. However, it should be noted that a smaller volume can reduce DNA elution efficiency and yield (the minimum should not be less than 25μL).