11. Put the adsorption column in a new centrifuge tube (self-provided), add 50-100 μL Buffer GE or sterile water dropwise to the middle of the adsorption column, leave it at room temperature for 2-5 minutes, and centrifuge at 12,000 rpm for 1 minute. Collect DNA solution and store at -20C.

Note: 1) If the downstream experiment is sensitive to pH or EDTA, it can be eluted with sterile water. The pH value of the eluent has a great influence on the elution efficiency. If water is used as the eluent, the pH value should be ensured to be between 7.0 and 8.5 (the pH value of the water can be adjusted to this range by NaOH). When the pH value is lower than 7.0 will reduce the elution efficiency.

2) Incubation at room temperature for 5 minutes prior to centrifugation can increase yield.

3) Elution with another 50-100 μL of Buffer GE or sterile water can increase the yield.

4) If you want to increase the final concentration of DNA, you can re-add the DNA eluate obtained in step 11 to the adsorption membrane and repeat step 11; you can increase the final concentration of DNA, but may reduce the total yield. If the amount of DNA is less than 1 μg, it is recommended to elute with 50 μl Buffer GE or sterile water.

5) DNA stored in water will be affected by acid hydrolysis. For long-term storage, it is recommended to elute with Buffer GE and store at -20C.

6) The residual micro-PCR inhibitor in the genomic DNA template may have an adverse effect on the PCR reaction, which can usually be solved by diluting the DNA 2-10 times.

Stool Genomic DNA kit

Cat. No. : CW2092S (50 preps)

Shipping and Storage : Storage at room temperature (15-30℃)

Components

Component	CW2092S (50 preps)
Buffer SW	60 mL
Buffer SL	60 mL
Buffer GL	50 mL
Buffer GW1 (concentrate)	2×13 mL
Buffer GW2 (concentrate)	15 mL
Buffer GE	15 mL
Spin Columns DM with Collection Tubes	50

Principle

This kit is suitable for extracting total DNA from stool samples, such as cells, bacteria, parasites and viruses, as well as PCR reaction inhibitors with high concentrations. This product can purify up to 300 mg of stool samples to obtain DNA fragments of 20-30 kb, and the purification process does not require toxic solvents such as phenol or chloroform, and ethanol precipitation. High-purity DNA fragments can be obtained within an hour.This kit uses a unique buffer system to efficiently bind DNA in the lysate to the adsorption column, while protein impurities in feces and other organic compounds that inhibit downstream reactions can flow through the membrane, inhibitors of PCR and enzymatic reactions, and residual Impurities can be effectively removed by a two-step washing step and finally eluted with a low-salt buffer or water to obtain high-purity DNA.The purified DNA can be directly used for enzyme digestion, PCR, Real-Time PCR, library construction, Southern Blot, molecular markers, etc.

Preparation and precautions before the experiment

1. Avoid repeated freezing and thawing of samples, otherwise the extracted DNA fragments will be smaller and the extraction yield will decrease.
2. Add absolute ethanol to Buffer GW1 and GW2 before the first use according to the instructions on the reagent bottle label.
3. Check Buffer SL and Buffer GL for crystallization or precipitation before use. If there is crystallization or precipitation, please re-dissolve Buffer SL and Buffer GL in a 56C water bath.
4. If the downstream experiment is sensitive to RNA contamination, you can add 4 μL DNase-Free RNase A (100 mg/ml) after adding Buffer SL. RNase A is not provided in this kit. If necessary, you can order it from our company separately. :CW0601S.

Procedure

1. Take 100-300 mg stool sample and place it in a centrifuge tube (self-provided).
2. Add 1 mL Buffer SW and then vortex for 3-5 minutes to disperse the sample evenly in the solution. Centrifuge at 12,000 rpm (~ 13,400 x g) for 1 minutes and then discard the supernatant.

3. Add 1mL Buffer SL and then vortex for 3-5 minutes to disperse the sample evenly in the solution. Bath in a water bath at 65C for 20 minutes, then ortex for 15 seconds every 5 minutes.

   Note: If RNA needs to be removed, add 4 μL 100 mg/mL RNase A solution (Cat. No.: CW0601S) after the above steps, shake and mix well, and leave at room temperature for 5-10 minutes.
4. Centrifuge at 12,000 rpm for 3 minutes and transfer the supernatant to a new centrifuge tube (self-provided).
5. Add an equal volume of Buffer GL to the supernatant, shake and mix well, and then place on ice for 5 minutes. Centrifuge at 12,000 rpm for 5 minutes.

   Note: The liquid may be transparent or turbid at this time, which will not affect the experiment.
6. Add the supernatant obtained in step 5 to the adsorption column (Spin Columns DM) that has been loaded into the collection tube. If the solution cannot be added at one time, it can be transferred in multiple times. Centrifuge at 12,000 rpm for 1 minute, and then discard the waste liquid in the collection tube, then put the adsorption column back into the collection tube.
7. Add 500 μL Buffer GW1 to the adsorption column (check whether absolute ethanol has been added before use), centrifuge at 12,000 rpm for 1 minute and then discard the waste liquid in the collection tube, then put the adsorption column back into the collection tube.
8. Repeat step 7.
9. Add 500 μL Buffer GW2 to the adsorption column (check whether absolute ethanol has been added before use), centrifuge at 12,000 rpm for 1 minute and then discard the waste liquid in the collection tube, and put the adsorption column back into the collection tube.
10. Centrifuge at 12,000 rpm for 2 minutes and then discard the waste in the collection tube. Allow the cartridge to dry at room temperature for several minutes.

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