

Universal Genomic DNA Extraction Kit

Cat No.: D2100

Package: 50T/100T

Storage: Keep it at room temperature, and the reinspection period is one year. (Note: RNase A, proteinase K is shipped as an accessory and stored at -20°C)

Kit content:

| Component | 50T | 100T | Storage |
|-------------------|----------|-----------|---------|
| RNase A | 100μL | 100μL×2 | -20°C |
| Proteinase K | 1 mL | 1 mL×2 | -20°C |
| Solution A | 25 mL | 50 mL | RT |
| Solution B | 25 mL | 50 mL | RT |
| Wash Buffer | 15 mL | 15 mL×2 | RT |
| Elution Buffer | 15 mL | 30 mL | RT |
| Adsorption Column | 50 units | 100 units | RT |
| Collecting Pipe | 50 units | 100 units | RT |
| Instruction | 1 | 1 | - |

Product description:

This kit is a universal type, suitable for extracting genomic DNA from soil, feces, insects, and other samples. For bacteria, fungi, insects and other samples have a good lysis effect, maximizing the retention of polymorphisms in biological DNA.

The DNA extracted using this kit has a large yield and good integrity, and can be directly used in various routine operations, including enzyme digestion, PCR, library construction, Southern hybridization and other experiments.

Operation steps (for reference only):

Before use, please add absolute ethanol to the rinse solution, and refer to the label on the bottle (add 45mL of absolute ethanol to each bottle separately). All centrifugation steps were performed at room temperature using a benchtop centrifuge.

1. Sample processing

1) Soil: weigh 0.1-0.3g (according to dry and wet) soil, put into the mortar, pour in the appropriate amount of liquid nitrogen, grind immediately, repeat 3 times, grind the soil particles into powder, add 500μL solution A, and oscillate to complete suspension.

2) Stool: weigh 0.1-0.3g of feces (according to dry and wet), add 500μL of solution A, and shake to complete suspension.





3) Insects: weigh 0.1-0.3g of insects, pour in an appropriate amount of liquid nitrogen, and immediately grind, repeat 3 times, grind the insects into powder, add 500 μ L of solution A, and shake to complete suspension.

4) Unknown samples, if fine, can directly weigh 0.1-0.3g (according to dry and wet) with 500 μ L solution A, grind 0.1-0.3g with liquid nitrogen, add 500 μ L solution A, and oscillate to complete suspension.

2. 2 μ L RNase A was added to the suspension, leaving it at 55 $^{\circ}$ C for 10min.

3. Add 20 μ L of proteinase K, mix thoroughly, and digested in 55 $^{\circ}$ C water bath for 30min. During digestion, the centrifuge tube was mixed several times and centrifuged at 12000rpm for 10min. The supernatant was transferred to a new centrifuge tube. If there is precipitation, it can be centrifuged again.

4. Add 500 μ L of solution B and mix well. If white precipitation appears, placed at 55 $^{\circ}$ C for 5min, the precipitation will disappear and will not affect the subsequent experiments. If the solution does not become clear, it means that the sample is not digested thoroughly, which may lead to the amount of extracted DNA less and impure, and may lead to the blocking of the column after the upper column, please increase the digestion time.

5. Add 500 μ L of anhydrous ethanol and mix well, flocculent precipitation may appear at this time, without affecting the extraction of DNA. Both solution and flocculent precipitation can be added to the adsorption column for 2min (add in two times, 700 μ L each time).

6. After centrifugation at 12000rpm for 2min, the waste solution was discarded and the adsorption column was placed into the collecting tube.

7. Add 600 μ L of rinse solution to the adsorption column (check whether absolute ethanol has been added before use), centrifuge at 12000rpm for 1min, discard the waste liquid and put the adsorption column into the collecting tube.

8. 600 μ L of rinse solution was added to the adsorption column, centrifuged at 12000rpm for 1min, the waste solution was discarded and the adsorption column was placed into the collecting tube

9. It was centrifuged at 12000rpm for 2min to place the adsorption column at room temperature or a 50 $^{\circ}$ C temperature box for several minutes to remove the residual rinse solution from the adsorption column, otherwise the ethanol in the rinse solution would affect subsequent experiments such as enzyme digestion, PCR, etc.

10. The adsorption column was placed in a clean centrifuge tube and 50-200 μ L of eluent preheated with 65 $^{\circ}$ C water bath was dropped to the center of the adsorption membrane, left at room temperature for 5min and centrifuged at 12000rpm for 2min.

11. The resulting eluate was added to the adsorption column for 2min at room temperature and at 12,000rpm for 2min to obtain high-quality genomic DNA.



Note:

1. Due to the different samples, the final DNA content and purity of the extraction also vary. Generally speaking, if the extracted DNA is not detected by electrophoresis, the PCR will have results and the sample will be as fresh as possible. Otherwise, it results in smaller extracted DNA fragments and decreased extraction amounts.
2. If the solution in the kit is precipitated, it can be redissolved in a 65°C water bath before use, without affecting the extraction effect.
3. If the sample is not thoroughly digested, the column may be blocked in the subsequent centrifugation step, and the centrifugation time can be appropriately extended appropriately.
4. The volume of the elution buffer should not be less than 50μL, the small volume will affect the recovery efficiency; the pH of the eluate also affects the elution efficiency, the pH value of the water can be NaOH to this range), and the pH value lower than 7.0 will reduce the elution efficiency; the DNA product should be stored at -20°C to prevent DNA degradation.
5. DNA concentration and purity detection (when the concentration is high): the size of the resulting genomic DNA fragment is related to the sample preservation time, the shear force during the operation and other factors. The recovered DNA fragments can be detected for concentration and purity by agarose gel electrophoresis and UV spectrophotometer. DNA should have a significant absorption peak at OD260 with an OD260 value of 1 equivalent to approximately 50μg/mL double-stranded DNA, 40μg/ mL single-stranded DNA. The OD260/ OD280 ratio should be 1.7-1.9. If the elution buffer is not used and deionized water is used, the ratio will be low because the pH and the presence of ions will affect the light absorption value, but does not indicate low purity.

Related products:

- D1010 6×DNA Loading Buffer*
- T1060 50×TAE Buffer*
- T1050 5×TBE Buffer*
- M1060 D2000 DNA Ladder*
- M1400 1kb DNA Ladder*
- G8142 GoldView Inucleic acid stain (5000×)*



