



Genorise Tissue DNA Extraction Kit 500

This kit is to isolate total DNA including genomic and mitochondrial DNA from 10 mg tissue and is for 500 applications. This kit can significantly improve quantity and quality of DNA and is much more cost-effective than the similar products.

Materials provided in the kit:

60 ml 5 x Lysis Solution
110 ml Protein Precipitation Solution
110 ml DNA Hydration Solution
1.0 ml 10 x DNA Sample Loading Buffer

Materials required but not provided in the kit:

Proteinase K, prepare at 20 mg/ml and store at -20°C
Isopropanol (2-propanol)/Ethanol

Protocol

Cell Lysis

1. Add 0.4 ml DNA water, 0.1 ml of 5 x Lysis Solution to 10 mg tissue in a 1.7 ml microtube, and vortex for 20 sec, and add 3 μ l Proteinase K solution (20 mg/ml), vortex for 20 sec.
2. Incubate at 55°C for 1 hr or overnight until the cell lysate becomes completely clear (disrupted).

Protein precipitation

1. Cool sample to room temperature by placing on ice for 1 min.
2. Add 0.2 ml of Protein Precipitation Solution to the lysate.
3. Vortex samples at high speed for 20 sec and place sample into an ice bath for 5 min.
4. Centrifuge at 15,000 x g for 5 min.

DNA Precipitation

1. Pour the supernatant containing DNA into a new 1.7 ml microcentrifuge tube.
2. Centrifuge at 15,000 x g for 5 min.
3. Pour the supernatant to a new microcentrifuge tube; repeat step 1 and 2 until no pellet is seen.
4. Pour the supernatant containing DNA into a new 1.5 ml microcentrifuge tube containing 0.5 ml 100% isopropanol (2-propanol).
5. Mix the samples by inverting gently 50 times and incubate at room temperature for 10 min.
6. Centrifuge at 13,000 x g for 5 min.
7. Pour off the supernatant and drain the tube briefly on clean absorbent paper. Add 1 ml of 70% ethanol and invert the tube several times to wash the DNA pellet.
8. Centrifuge at 13,000 x g for 5 min, carefully pour off the ethanol and do not lose the DNA pellet.
9. Invert and drain the tube on clean absorbent paper, completely remove the remaining liquid by a pipette, and finally allow air dry for 5 min.

DNA Hydration

1. Add 50 μ l DNA Hydration Solution.
2. Resuspend the DNA by a pipette for 5 times and rehydrate the DNA by incubation for 10 min at room temperature.
3. Vortex briefly and pulse spin before use, and store at -20°C.

Note

1. Following DNA precipitation, if you see a big pellet with a color other than white, we recommend you repeat this protocol with a start volume of 0.5 ml of DNA isolate and 1 hr cell lysis.
2. Do not air dry DNA pellet more than 5 min.