Magnetic Fast Tissue DNA Kit

Cat.No.: HYC284

Product Description

This kit delivers a simple, rapid and high-efficiency method for DNA isolation from fresh or frozen tissues. After tissue lysis, DNA binds to the surface of silica-coated magnetic beads. Following washing steps, highly pure DNA is eluted in TB buffer or deionized water. Purified DNA can be directly applied to PCR, Real-time PCR, SNP genotyping, STR genotyping, next-generation sequencing, pharmacogenomic research and other downstream assays. In addition to rapid DNA extraction from animal tissues, this kit is also compatible with samples including blood, cultured cells and bacteria.

Components

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Buffer ATL	25 mL
Buffer AL	36 mL
Buffer MB	70 mL
Buffer DW	70 mL
Buffer TB	15 mL
Proteinase K (20 mg/mL)	2×1.25 mL
RNase A (100 mg/mL)	0.6 mL
Magnetic Microspheres N	4×1.25 mL

Storage

All components can be stably stored in a dry place at room temperature (15-30℃).

Required Reagents

Absolute Ethanol

Instructions

Preparations

1. Inspect Buffer ATL and Buffer AL for crystals or precipitates prior to use. If solids are observed, dissolve the buffers completely by incubation in a 56 °C water bath.
2. Preheat a water bath or thermomixer to 56 °C before starting experiments.
3. Magnetic Microspheres N must not be frozen or subjected to high-speed centrifugation; improper handling may cause irreversible damage to Magnetic Microspheres N. Vortex Magnetic Microspheres N thoroughly to achieve uniform suspension each time before use.

Starting Sample Amounts

- Animal tissue: 25–30 mg (spleen tissue should be less than 10 mg). A 2 mm cubic tissue piece (volume: 8 mm^3) weighs approximately 10–15 mg.
- Blood: 200–300 µL for mammalian blood samples; around 10 µL for avian and amphibian blood samples.
- Cultured mammalian cells: 1×10^6 cells.
- Bacteria: 1×10^9 cells.

Procedures (Animal Tissue)

This protocol is applicable to fresh and frozen tissue samples. The yield and quality of purified DNA depend on tissue type, source and storage conditions. For cell, bacterial and other samples, pellet the material using the recommended starting amount, then follow the animal tissue extraction workflow. Gram-positive bacteria require pre-treatment with lysozyme.

1. Weigh out 25–30 mg animal tissue (spleen tissue weight must not exceed 10 mg). Grind tissue into fine powder with liquid nitrogen or cut into small pieces with a scalpel, then transfer into a 2 mL centrifuge tube pre-filled with 220 µL Buffer ATL. Vortex thoroughly and perform brief centrifugation.
2. Add 20 µL Proteinase K, vortex to mix evenly and spin briefly.
3. Incubate at 56 °C with shaking at 900 rpm for 30 min until complete tissue digestion. Spin briefly to collect liquid droplets on the inner wall of the tube cap.
4. Centrifuge at 14,000 × g for 2 min, transfer 200 µL of the lysate supernatant into a new 2 mL centrifuge tube.

5. Add 5 μL RNase A, mix by gentle vortexing, and incubate at room temperature for 10 min.
6. Add 150 μL Buffer AL, mix with gentle vortexing and spin briefly.
7. Add 280 μL isopropanol and 40 μL Magnetic Microspheres N. Gently vortex for 10 s and spin briefly. Shake at 1200 rpm at room temperature for 10 min, then perform brief centrifugation to remove liquid from the tube cap.
8. Place the tube on a magnetic rack for 1 min (★ until the solution turns clear). Discard supernatant without touching the magnetic beads.
9. Add 650 μL Buffer MB to the tube, gently vortex for 10 s to resuspend beads, then shake at 1200 rpm under room temperature for 3 min. Spin briefly to clear liquid from the cap. Place the tube on the magnetic rack for 1 min (★ until the solution turns clear) and discard supernatant; avoid contact with beads.
10. Add 650 μL Buffer DW to the tube, gently vortex for 10 s to resuspend beads, then shake at 1200 rpm under room temperature for 3 min. Spin briefly to clear liquid from the cap. Place the tube on the magnetic rack for 1 min (★ until the solution turns clear) and discard supernatant; avoid contact with beads.
11. Add 650 μL self-supplied 75% ethanol, gently vortex for 10 s to resuspend beads, then shake at 1200 rpm under room temperature for 3 min. Spin briefly to clear liquid from the cap. Place the tube on the magnetic rack for 1 min (★ until the solution turns clear) and discard supernatant; avoid contact with beads.
12. Repeat Step 11 once.
13. Air-dry the tube with all supernatant removed at room temperature for 3–5 min until beads appear matte black with no surface gloss. Add 80–100 μL Buffer TB and gently vortex for 10 s to fully resuspend beads. Incubate at 56 °C with shaking at 1200 rpm for 10 min, then spin briefly to clear liquid from the cap. Set the tube on the magnetic rack for 1 min, transfer the supernatant containing purified DNA into a new centrifuge tube for subsequent use. Purified DNA can be stored long-term at −20 °C.

Procedures (Animal Blood)

This protocol is suitable for fresh or frozen anticoagulated whole blood stabilized with EDTA or sodium citrate. Thaw frozen blood rapidly at 37 °C and mix thoroughly by gentle vortexing;

equilibrate samples to room temperature before starting the experiment. The yield and quality of purified DNA vary with blood storage conditions; fresh blood delivers superior results.

1. Add 20 μL Proteinase K and 200 μL blood to a 1.5 mL centrifuge tube, then mix via gentle vortexing.
2. Spin briefly, add 300 μL Buffer AL and 5 μL RNase A, vortex gently for 10 seconds, and spin briefly. Incubate at 56 °C with shaking at 900 rpm for 15 min.
3. After incubation, spin briefly. Add 300 μL isopropanol and 20 μL Magnetic Microspheres N, vortex gently for 10 s, then shake at 900 rpm at room temperature for 5 min. Spin briefly to remove residual liquid on the tube cap.
4. Place the tube on a magnetic rack for 1 min (★ until the solution turns clear). Carefully discard supernatant without touching the magnetic beads.
5. Add 650 μL Buffer MB to the tube, vortex gently for 10 s to resuspend beads, then shake at 1200 rpm at room temperature for 3 min. Spin briefly to clear liquid from the tube cap. Set the tube on the magnetic rack for 1 min (★ until the solution clarifies) and discard supernatant; avoid bead contact.
6. Add 650 μL Buffer DW to the tube, vortex gently for 10 s to resuspend beads, then shake at 1200 rpm at room temperature for 3 min. Spin briefly to clear liquid from the tube cap. Set the tube on the magnetic rack for 1 min (★ until the solution clarifies) and discard supernatant; avoid bead contact.
7. Add 650 μL self-supplied 75% ethanol to the tube, vortex gently for 10 s to resuspend beads, then shake at 1200 rpm at room temperature for 3 min. Spin briefly to clear liquid from the tube cap. Set the tube on the magnetic rack for 1 min (★ until the solution clarifies) and discard supernatant; avoid bead contact.
8. Repeat Step 7 once.
9. Air-dry the tube with all supernatant removed at room temperature for 3–5 min until beads appear matte black with no surface gloss. Add 80–100 μL Buffer TB and vortex gently for 10 s to fully resuspend beads. Incubate at 56 °C with shaking at 1200 rpm for 10 min, then spin briefly to clear liquid from the tube cap. Place the tube on the magnetic rack for 1 min, transfer the supernatant containing purified DNA into a new centrifuge tube for subsequent use. Purified DNA can be stored long-term at −20 °C.