Micro Sample Genomic DNA Kit

Cat. No.: M1522624 | Pack size: 50T | Storage: Room temperature; −20 °C

Product Description

This kit uses a spin column that specifically binds nucleic acids and a unique buffer system. After sample lysis, DNA binds to the silica membrane under high-salt conditions and is eluted from the membrane under low-salt, high-pH conditions. This kit is suitable for extracting genomic DNA from small amounts of samples such as low-volume blood, dried blood spots, serum/plasma, micro-tissues, mouthwash, hair, and microdissected tissues. The obtained genomic DNA can be directly used in molecular biology experiments such as PCR templates, restriction enzyme digestion, and hybridization.

Contents & Storage

Product No.	Component	Appearance	50T	Storage
M1522624A	Buffer UA	Liquid	15 mL	RT
M1522624B	Buffer BDL	Liquid	15 mL	RT
M1522624C	Buffer PW1	Liquid	17 mL	RT
M1522624D	Buffer PW2	Liquid	15 mL	RT
M1522624E	Buffer EB	Liquid	15 mL	RT
M1522624F	Proteinase K	Liquid	1.2 mL	RT
M1522624G	GHT Spin Column (with 2 mL collection tube)	—	1 EA × 50	RT
M1522624H	Trace RNA Enhancer	Liquid	280 µL	−20 °C

Shelf life: The kit can be stored under dry conditions at room temperature for 15 months.

Note: The Trace RNA Enhancer is shipped with ice packs; it can be stored at 4 °C for 1 month and at −20 °C for long-term storage.

Precautions

The user needs to supply absolute ethanol, 1 M DTT (for hair follicle extraction), RNase A (10 mg/mL), centrifuge tubes, sterile tubes, a hole punch, and other auxiliary tools.

Bring all samples to room temperature before use. If the room temperature exceeds 25 °C, pre-cool the absolute ethanol on ice to avoid affecting the results.

To maximize DNA yield from trace samples, this kit includes a Trace RNA Enhancer. Because the Enhancer itself is a small nucleic acid, the measured OD_{260} value of the genomic DNA will be higher than the actual value. It is recommended to directly test the obtained genomic DNA by PCR.

Before use, add absolute ethanol to Buffer PW1 and Buffer PW2. Buffer UA may precipitate at low temperatures; redissolve in a 37 °C water bath and shake well before use.

Procedure

Before use, add 22 mL of absolute ethanol to Buffer PW1 and 60 mL of absolute ethanol to Buffer PW2.

This kit provides dedicated extraction protocols for different trace sample types. Follow the respective steps according to the sample type. After completing the corresponding steps, all samples proceed to step 8.

1. Low-volume blood samples

(1) Transfer 1–100 µL of blood to a 1.5 mL centrifuge tube. If the blood volume is less than 100 µL, bring the volume to 100 µL with Buffer UA.

(2) If RNA removal is required, add 5 µL of RNase A (10 mg/mL) solution (user-supplied), vortex to mix, and let stand at room temperature for 5 min.

(3) Add 10 µL of Proteinase K and vortex to ensure thorough contact between the enzyme and the sample.

(4) Add 100 µL of Buffer BDL, mix thoroughly by inversion, and incubate at 56 °C for 10 min, mixing the sample 2–3 times during incubation. After incubation, briefly centrifuge to collect the solution from the cap wall, avoiding reagent residue.

Note: *If the blood sample volume is <10 µL, add 5 µL of Trace RNA Enhancer to the 100 µL of Buffer BDL. White precipitate may form upon addition of Buffer BDL, but usually disappears during incubation at 56 °C and does not affect subsequent steps. If the solution does not become clear, cell lysis is incomplete, which may lead to reduced DNA yield and purity.*

(5) Add 50 µL of absolute ethanol, mix gently by inversion, and let stand at room temperature for 3 min. Briefly centrifuge to remove drops from the cap wall.

(6) Proceed to step 8.

2. Dried blood spots

(1) Using a hole punch or scissors, take three 3×3 mm pieces of dried blood spot sample and place into a 1.5 mL centrifuge tube.

(2) Add 180 µL of Buffer UA to completely immerse the sample.

(3) Add 20 µL of Proteinase K, vortex to mix, and incubate at 56 °C for 60 min, inverting to mix every 10 min to promote lysis.

(4) Add 200 µL of Buffer BDL, vortex to mix, and incubate at 70 °C for 10 min, vortexing every 3 min. After incubation, briefly centrifuge to collect the solution from the cap wall.

Note: *White precipitate may form upon addition of Buffer BDL, but usually disappears during incubation at 70 °C. If the solution does not become clear, lysis is incomplete, which may lead to low DNA yield and purity.*

(5) Add 200 µL of absolute ethanol, mix gently by inversion, and let stand at room temperature for 5 min. Briefly centrifuge to remove drops from the cap wall.

(6) Proceed to step 8.

3. Micro-tissue samples

(1) Weigh no more than 10 mg of animal tissue and transfer to a 1.5 mL centrifuge tube pre-filled with 180 µL of Buffer UA.

(2) Add 20 µL of Proteinase K, vortex to mix, and incubate at 56 °C for 30–60 min, inverting to mix every 15 min to ensure complete tissue lysis.

(3) Add 200 µL of Buffer BDL and 5 µL of Trace RNA Enhancer, vortex to mix, and incubate at 70 °C for 10 min, vortexing every 3 min. After the solution becomes clear, briefly centrifuge to collect the solution from the cap wall.

(4) Add 200 µL of absolute ethanol, mix gently by inversion, and let stand at room temperature for 5 min. Briefly centrifuge to remove drops from the cap wall.

(5) Proceed to step 8.

4. Microdissected samples (including formalin-fixed microdissected samples)

(1) Add 15 µL of Buffer UA to a 0.2 mL centrifuge tube and place the microdissected sample into the tube.

(2) Add 10 µL of Proteinase K and vortex to ensure thorough contact.

(3) Incubate at 56 °C for 3 h (formalin-fixed samples require 16 h) until the sample is completely lysed, mixing occasionally.

(4) Add 25 µL of Buffer UA and mix, then add 50 µL of Buffer BDL and 5 µL of Trace RNA Enhancer, vortex to mix. Briefly centrifuge to collect the solution from the cap wall.

(5) Add 50 µL of absolute ethanol, mix gently by inversion, and let stand at room temperature for 5 min.

(6) Proceed to step 8.

5. Forensic materials (hair, body fluid-stained materials, nails)

(1) Sample processing

Hair: Take 1 cm of hair from the root (including the follicle) and place into a 1.5 mL centrifuge tube. Add 250 µL of Buffer UA, 20 µL of Proteinase K, and 20 µL of 1 M DTT solution. Vortex to mix. Proceed to step (2).

Body fluid-stained material: Take approximately 0.5 cm^3 of material stained with blood, saliva, or semen, cut into small pieces, and place into a 1.5 mL centrifuge tube. Add 300 µL of Buffer UA and 20 µL of Proteinase K. Vortex to mix. Proceed to step (2). ***Note:*** *For semen samples, additionally add 20 µL of 1 M DTT solution.*

Nails: Cut nails into small pieces and place into a 1.5 mL centrifuge tube. Add 300 µL of Buffer UA, 20 µL of Proteinase K, and 20 µL of 1 M DTT solution. Vortex to mix. Proceed to step (2).

(2) Incubate at 56 °C until the sample is completely lysed, inverting to mix every 10 min. ***Note:*** *Hair usually lyses in 60 min; tough materials such as nails may require overnight lysis.*

(3) Add 300 µL of Buffer BDL and 5 µL of Trace RNA Enhancer, vortex to mix, and incubate at 56 °C for 10 min, vortexing every 3 min. Briefly centrifuge to collect the solution from the cap wall.

(4) Add 300 µL of absolute ethanol, mix gently by inversion, and let stand at room temperature for 3 min.

(5) Proceed to step 8.

6. Circulating cell-free nucleic acid extraction from serum/plasma

(1) Transfer 100–200 µL of serum/plasma to a 1.5 mL centrifuge tube. If the volume is less than 100 µL, bring to volume with Buffer UA.

(2) Add 20 µL of Proteinase K and vortex to mix.

(3) Add 200 µL of Buffer BDL, mix thoroughly by inversion, and incubate at 56 °C for 10 min, mixing the sample 2–3 times during incubation. Briefly centrifuge to collect the solution from the cap wall. ***Note:*** *If the serum/plasma volume is <50 µL, add 5 µL of Trace RNA Enhancer to the 200 µL of Buffer BDL. White precipitate may form upon addition of Buffer BDL, but usually disappears during incubation at 56 °C.*

(4) Add 200 µL of absolute ethanol, mix gently by inversion, and let stand at room temperature for 5 min.

(5) Proceed to step 8.

7. Genomic DNA extraction from mouthwash

(1) Add 10–20 mL of mouthwash sample to a 50 mL sterile tube. Centrifuge at 800 rpm for 5 min. Carefully pour off the supernatant, retaining the pellet at the bottom.

(2) Add 200 µL of Buffer UA to the pellet and resuspend thoroughly. Transfer the entire suspension to a 1.5 mL centrifuge tube.

(3) Add 20 µL of Proteinase K and vortex to mix. Incubate at 56 °C for 60 min, vortexing every 15 min to promote lysis of the pellet.

(4) Add 200 µL of Buffer BDL and 5 µL of Trace RNA Enhancer, mix thoroughly by inversion, and incubate at 70 °C for 10 min, vortexing every 3 min. After the solution becomes clear, briefly centrifuge to collect the solution from the cap wall. ***Note:*** *White precipitate may form upon addition of Buffer BDL, but usually disappears during incubation at 70 °C. If the solution does not become clear, lysis is incomplete, which may lead to low yield and purity.*

(5) Add 200 µL of absolute ethanol, mix thoroughly by inversion, and briefly centrifuge to remove drops from the cap wall.

(6) Proceed to step 8.

8. Transfer the solution obtained from the respective sample steps above into a GHT spin column (placed in the provided 2 mL collection tube). Centrifuge at 12,000 rpm (13,680 × g) for 30 sec. Discard the flow-through and return the spin column to the collection tube.

9. Add 700 µL of Buffer PW1 (with ethanol pre-added) to the spin column. Centrifuge at 12,000 rpm (13,680 × g) for 30 sec. Discard the flow-through and return the spin column to the collection tube.

10. Add 700 µL of Buffer PW2 (with ethanol pre-added) to the spin column. Centrifuge at 12,000 rpm (13,680 × g) for 30 sec. Discard the flow-through and return the spin column to the collection tube.

11. Repeat step 10 to further remove residual salts, proteins, and other impurities on the spin column, thereby increasing DNA purity.

12. Centrifuge the spin column together with the collection tube at 12,000 rpm (13,680 × g) for 2 min to completely remove residual wash buffer and impurities (this avoids residual ethanol affecting subsequent DNA elution and experiments).

13. DNA elution

Transfer the spin column to a new 1.5 mL centrifuge tube. Open the cap and let stand for 2 min to allow residual ethanol to evaporate completely. Slowly add 20 µL of Buffer EB dropwise to the center of the membrane without touching it. Let stand at room temperature for 1 min. Centrifuge at 12,000 rpm (13,680 × g) for 30 sec. Collect the DNA solution. The DNA can be used immediately or stored at −20 °C for long-term preservation.

Note: *To increase DNA yield, reload the first eluate onto the spin column and repeat the elution, or pre-heat Buffer EB to 65 °C before use.*

Storage and Shipping

Item	Description
Storage	Room temperature; store at −20 °C
Shipped In	Wet ice
Stability and Storage	Each component has a shelf life of 15 months under corresponding storage conditions. M1522624A, M1522624B, M1522624C, M1522624D, M1522624E, M1522624F, M1522624G: Store at room temperature long term (15 months). M1522624H: Store at −20 °C long term (15 months).

Quality Control

QC Item	Method	Acceptable Range
Component completeness	Visual inspection on receipt	All components and counts match the contents table; lot number and expiry date are legible.
Genomic DNA recovery	Extraction from representative samples followed by fluorometric or spectrophotometric assessment	Recovered DNA should be suitable for the intended downstream application; yield varies with sample type, input amount, and sample condition.
DNA purity and usability	Spectrophotometry and downstream PCR verification as applicable	Eluted DNA should be compatible with PCR, restriction enzyme digestion, or hybridization after user validation.

Troubleshooting

Issue	Possible Causes	Corrective Action
Low DNA yield	• Insufficient sample lysis • Incomplete ethanol addition to PW1/PW2 • Elution volume or elution contact time too low	• Extend lysis time for tough samples and mix thoroughly during incubation • Confirm that ethanol was added before first use • Reload the eluate for a second elution or pre-heat Buffer EB to 65 °C
Low DNA purity or PCR inhibition	• Carryover of wash buffer or ethanol • Excess sample input or incomplete washing	• Perform the recommended drying spin completely • Reduce sample input and repeat wash steps as needed
Spin column clogging or slow flow	• Excess tissue or insoluble debris • Incomplete lysis or particulate material transferred to the column	• Use no more than the recommended sample amount • Ensure complete lysis and avoid transferring visible debris where possible

Recommended Applications

PCR templates · restriction enzyme digestion · hybridization

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NORTH AMERICAN SALES, SUPPORT & GENERAL INQUIRIES Aladdin Scientific Corporation 14078 Meridian Parkway, Riverside, CA 92518, USA Phone: 1-833-552-7181 Sales: sales@aladdinsci.com Customer Service: custserv@aladdinsci.com	EU SALES, LOGISTICS & LOCALIZED SUPPORT Aladdin Biochem Deutschland GmbH Westring 2, 33142 Büren, Germany Phone: +02951 9383958 Support: support.eu@aladdinsci.com

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