ONEOMICS | YOUR GENOMICS PARTNER

ONEMag™ Universal DNA Extraction Kit

(ONE-ULDNA-50)

Intended Use:

The **ONEMag™ Universal DNA Extraction Kit** is intended for the isolation of high-quality genomic DNA from a wide range of human clinical samples, including blood, saliva, urine, vaginal swab, tissue, and faecal specimens. The purified DNA is suitable for downstream molecular applications such as PCR, qPCR, and next-generation sequencing (NGS). This kit is for research use only.

Principle:

The **ONEMag™ Universal DNA Extraction Kit** utilizes silica-coated magnetic bead technology for the isolation of high-quality genomic DNA from diverse human clinical specimens.

Samples are lysed using a proprietary chaotropic lysis buffer to disrupt cell membranes and release genomic DNA. Under optimized binding conditions, DNA selectively binds to the magnetic beads, while proteins, polysaccharides, inhibitors, and other contaminants remain in solution. The magnetic beads are separated using a magnetic field, washed to remove residual impurities, and DNA is finally eluted in a low-salt buffer.

Kit Contents:

Components	Quantity (50 Reactions)
CL Buffer	40 mL
BB Buffer	30 mL
Magnetic Bead	3 mL
BW Buffer (Concentrate)	15 mL
BE Buffer	3 mL

Material's/Equipment's required but not Supplied:

- Ethanol (96–100%), Molecular Grade
- Micropipettes and sterile tips
- 1.5 mL microcentrifuge tubes
- Heating block or Water bath
- Vortex mixer
- Magnetic stand for 1.5/2.0 mL tubes
- Proteinase K

Storage Conditions:

All components of the **ONEMag™ Universal DNA Extraction Kit** should be stored at room temperature (~25°C). The kit is stable until the expiration date printed on the product label when stored under recommended conditions.

Preparation of Buffers:

Preparation of BW Buffer

BW Buffer is supplied as a **concentrate** and must be diluted with ethanol before use.

Component	Volume for 50 Reactions
BW Buffer (Concentrate)	15 mL
Ethanol (96-100%)	45 mL
Final Volume	60 mL

Mix thoroughly and store at room temperature.

Safety and Precautions:

This kit is for research use only and is not intended for diagnostic or therapeutic applications. All human-derived samples should be considered potentially infectious and handled in accordance with appropriate biosafety guidelines. Wear suitable personal protective equipment, including laboratory coat, gloves, and eye protection, when handling samples and reagents. Kit reagents may contain chaotropic salts that are harmful if swallowed, inhaled, or in contact with skin or eyes; avoid direct contact and rinse immediately with water in case of exposure.

A. Whole Blood DNA Extraction Protocol:

1. Add 500 µL CL Buffer to 100 µL whole blood sample and vortex thoroughly.
2. Add Proteinase K to a final concentration of 0.3 mg/mL. *(Proteinase K is not supplied with this kit; for example, add 8 µL of 20 mg/mL Proteinase K to 500 µL lysis buffer.)*
3. Incubate at 60 °C for 15 minutes.
4. Invert the tube several times during incubation to mix the sample.
5. Centrifuge at 6,000 rpm for 10 minutes and transfer 400 µL of lysate to a new 1.5 mL tube.
6. Add 400 µL of BB buffer and vortex well.
7. Add 20 µL of magnetic beads and vortex thoroughly.
8. Incubate at room temperature for 5 minutes.
9. Place the tube on a magnetic stand and allow the beads to separate for 30 seconds.
10. Carefully discard the supernatant without disturbing the beads.
11. Add 500 µL of BW buffer and vortex well.
12. Place the tube on the magnetic stand for 30 seconds.
13. Carefully discard the supernatant.
14. Repeat the wash step once more (Steps 11–13).
15. Remove any residual buffer and air-dry the beads for 1 minute. Do not over-dry.
16. Add 50 µL of BE buffer, vortex briefly, and incubate at room temperature for 5 minutes.
17. Place the tube on the magnetic stand for 1 minute, then transfer the eluted DNA to a new tube and store at –20 °C

B. Saliva DNA Extraction Protocol:

1. Transfer 500 µL of Saliva sample into a 1.5 mL microcentrifuge tube and centrifuge at 10,000 rpm for 5 minutes.
2. Carefully discard 400 µL of the supernatant, then add 500 µL CL Buffer and vortex thoroughly.
3. Add Proteinase K to a final concentration of 0.4 mg/mL. *(Proteinase K is not supplied with this kit; for example, add 10 µL of 20 mg/mL Proteinase K to 500 µL lysis buffer.)*
4. Incubate at 65 °C for 15 minutes.
5. Invert the tube several times during incubation to mix the sample.
6. Centrifuge at 6,000 rpm for 10 minutes and transfer 500 µL of lysate to a new 1.5 mL tube.
7. Add 500 µL of BB buffer and vortex well.
8. Add 20 µL of magnetic beads and vortex thoroughly.
9. Incubate at room temperature for 5 minutes.
10. Place the tube on a magnetic stand and allow the beads to separate for 30 seconds.
11. Carefully discard the supernatant without disturbing the beads.
12. Add 500 µL of BW buffer and vortex well.
13. Place the tube on the magnetic stand for 30 seconds.
14. Carefully discard the supernatant.
15. Repeat the wash step once more (Steps 12–14).
16. Remove any residual buffer and air-dry the beads for 1 minute. Do not over-dry.
17. Add 50 µL of BE buffer, vortex briefly, and incubate at room temperature for 5 minutes.
18. Place the tube on the magnetic stand for 1 minute, then transfer the eluted DNA to a new tube and store at –20 °C.

C. Urine DNA Extraction Protocol:

1. Transfer 1000 µL of Urine sample into a 1.5 mL microcentrifuge tube and centrifuge at 10,000 rpm for 8 minutes.
2. Carefully discard 900 µL of the supernatant, then add 500 µL CL Buffer and vortex thoroughly.
3. Add Proteinase K to a final concentration of 0.4 mg/mL. *(Proteinase K is not supplied with this kit; for example, add 10 µL of 20 mg/mL Proteinase K to 500 µL lysis buffer.)*
4. Incubate at 65 °C for 15 minutes.
5. Invert the tube several times during incubation to mix the sample.
6. Centrifuge at 6,000 rpm for 10 minutes and transfer 400 µL of lysate to a new 1.5 mL tube.
7. Add 400 µL of BB buffer and vortex well.
8. Add 20 µL of magnetic beads and vortex thoroughly.
9. Incubate at room temperature for 5 minutes.
10. Place the tube on a magnetic stand and allow the beads to separate for 30 seconds.
11. Carefully discard the supernatant without disturbing the beads.
12. Add 500 µL of BW buffer and vortex well.
13. Place the tube on the magnetic stand for 30 seconds.
14. Carefully discard the supernatant.
15. Repeat the wash step once more (Steps 12–14).
16. Remove any residual buffer and air-dry the beads for 1 minute. Do not over-dry.
17. Add 50 µL of BE buffer, vortex briefly, and incubate at room temperature for 5 minutes.
18. Place the tube on the magnetic stand for 1 minute, then transfer the eluted DNA to a new tube and store at –20 °C.

D. Vaginal swab DNA Extraction Protocol:

1. Immediately after collection, place each swab into a sterile tube containing transport medium (e.g., PBS, TE buffer, or a specialized DNA/RNA stabilization solution). Vortex thoroughly for 2 minutes to mix the sample.
2. Transfer 500 µL of the medium into a 1.5 mL microcentrifuge tube and centrifuge at 10,000 rpm for 5 minutes.
3. Carefully remove 400 µL of the supernatant. Add 500 µL of CL Buffer and vortex thoroughly to resuspend the pellet.
4. Add Proteinase K to a final concentration of 0.4 mg/mL. *(Proteinase K is not supplied with this kit; for example, add 10 µL of 20 mg/mL Proteinase K to 500 µL lysis buffer.)*
5. Incubate at 65 °C for 15 minutes.
6. Invert the tube several times during incubation to mix the sample.
7. Centrifuge at 6,000 rpm for 10 minutes and transfer 400 µL of lysate to a new 1.5 mL tube.
8. Add 400 µL of BB buffer and vortex well.
9. Add 20 µL of magnetic beads and vortex thoroughly.
10. Incubate at room temperature for 5 minutes.
11. Place the tube on a magnetic stand and allow the beads to separate for 30 seconds.
12. Carefully discard the supernatant without disturbing the beads.
13. Add 500 µL of BW buffer and vortex well.
14. Place the tube on the magnetic stand for 30 seconds.
15. Carefully discard the supernatant.
16. Repeat the wash step once more (Steps 13–15).
17. Remove any residual buffer and air-dry the beads for 1 minute. Do not over-dry.
18. Add 50 µL of BE buffer, vortex briefly, and incubate at room temperature for 5 minutes.
19. Place the tube on the magnetic stand for 1 minute, then transfer the eluted DNA to a new tube and store at –20 °C.

E. Faecal DNA Extraction Protocol:

1. Transfer up to 100 mg of faecal sample into a 1.5 mL microcentrifuge tube**.** Add 600 µL CL Buffer and vortex thoroughly.
2. Add Proteinase K to a final concentration of 0.3 mg/mL. *(Proteinase K is not supplied with this kit; for example, add 9 µL of 20 mg/mL Proteinase K to 600 µL lysis buffer.)*
3. Incubate at 65 °C for 20 minutes.
4. During incubation, invert the tube several times to ensure proper mixing.
5. Centrifuge at 7,000 rpm for 10 minutes and transfer 400 µL of the clear lysate to a new 1.5 mL tube.
6. Add 400 µL BB Buffer and vortex well.
7. Add 20 µL magnetic beads and vortex thoroughly.
8. Incubate at room temperature for 5 minutes**.**
9. Place the tube on a magnetic stand and allow the beads to separate for 30 seconds**.**
10. Carefully discard the supernatant without disturbing the bead pellet.
11. Add 500 µL BW Buffer and vortex to resuspend the beads.
12. Place the tube on the magnetic stand for 30 seconds**.**
13. Carefully discard the supernatant.
14. Repeat the wash step once more (Steps 11–13).
15. Remove any residual buffer and air-dry the beads for 1 minute**.** Do not over-dry.
16. Add 50 µL BE Buffer**,** vortex briefly, and incubate at room temperature for 5 minutes**.**
17. Place the tube on the magnetic stand for 1 minute**,** transfer the eluted DNA to a new tube, and store at –20 °C**.**

F. Tissue DNA Extraction Protocol:

1. Transfer up to 50 mg of tissue into a 1.5 mL microcentrifuge tube. Add 600 µL CL Buffer and homogenize thoroughly.
2. Add Proteinase K to a final concentration of 0.4 mg/mL. *(Proteinase K is not supplied with this kit; for example, add 12 µL of 20 mg/mL Proteinase K to 600 µL lysis buffer.)*
3. Incubate at 65 °C for 20 minutes.
4. During incubation, invert the tube several times to ensure proper mixing.
5. Centrifuge at 6,000 rpm for 10 minutes and transfer 400 µL of the clear lysate to a new 1.5 mL tube.
6. Add 400 µL BB Buffer and vortex well.
7. Add 20 µL magnetic beads and vortex thoroughly.
8. Incubate at room temperature for 5 minutes.
9. Place the tube on a magnetic stand and allow the beads to separate for 30 seconds.
10. Carefully discard the supernatant without disturbing the bead pellet.
11. Add 500 µL BW Buffer and vortex to resuspend the beads.
12. Place the tube on the magnetic stand for 30 seconds.
13. Carefully discard the supernatant.
14. Repeat the wash step once more (Steps 11–13).
15. Remove any residual buffer and air-dry the beads for 1 minute. Do not over-dry.
16. Add 50 µL BE Buffer, vortex briefly, and incubate at room temperature for 5 minutes.
17. Place the tube on the magnetic stand for 1 minute, transfer the eluted DNA to a new tube, and store at –20 °C.

Troubleshooting Guide:

Problem Observed	Possible Cause	Recommended Solution
Low DNA yield	Incomplete sample lysis	Ensure thorough vortexing or homogenization during the lysis step. Verify correct incubation temperature and time. Increase incubation time by 5–10 minutes for difficult samples (faecal or tissue).
	Insufficient Proteinase K activity	Confirm correct Proteinase K concentration and that the enzyme is active. Avoid repeated freeze–thaw cycles of Proteinase K.
	Low starting material	Increase input sample volume or mass within recommended limits.
	Inefficient DNA binding	Ensure BB Buffer is added at the correct volume and mixed thoroughly. Vortex immediately after adding magnetic beads.
Poor DNA purity (A260/280 or A260/230 low)	Incomplete removal of wash buffer	After the final wash, ensure complete removal of BW Buffer before elution. Briefly spin down and re-place on magnetic stand if needed.
	Beads over-dried	Do not air-dry beads for more than 1 minute. Over-drying reduces elution efficiency and purity.
	Carryover of inhibitors (especially faecal samples)	Carefully transfer only clear lysate after centrifugation. Avoid disturbing debris or pellet.
DNA does not amplify in PCR/qPCR	Presence of PCR inhibitors	Perform an additional BW wash if inhibitors are suspected. Dilute the eluted DNA (1:5 or 1:10) before PCR.
	Low DNA concentration	Quantify DNA before downstream use. Increase elution efficiency by warming BE Buffer to 55–60 °C before elution.
Magnetic beads not separating properly	Magnetic stand not suitable	Ensure the magnetic stand is compatible with 1.5/2.0 mL tubes and strong enough for bead separation.
	Insufficient separation time	Increase magnetic separation time to 1–2 minutes if beads remain suspended.
Bead carryover in eluted DNA	Disturbance of beads during elution	When transferring eluted DNA, avoid touching the bead pellet. Place the tube back on the magnetic stand briefly if beads are visible.
DNA degradation	Prolonged storage of samples before extraction	Process samples promptly or store appropriately (e.g., blood at 4 °C short-term, −20 °C long-term). Avoid repeated freeze–thaw cycles.

Variable yields between samples	Inconsistent sample processing	Standardize sample input volume/mass and ensure uniform vortexing, incubation, and centrifugation conditions across samples.