



MAGN^{XT}

Cell free DNA Extraction Kit

REF

G2M810003

| S. No. | Product Name | SKU Code | Pack Size |
|--------|-------------------------------------|----------------|-----------|
| 1. | MagNXT Cell free DNA Extraction Kit | G2M810003-50T | 50T |
| 2. | MagNXT Cell free DNA Extraction Kit | G2M810003-250T | 250T |



MADE IN INDIA



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Intended Use

MagNXT Cell free DNA Extraction Kit is an *in-vitro* diagnostic test kit, intended for isolation and purification of Cell free DNA from clinical samples such as, serum, plasma. MagNXT Cell free DNA Extraction Kit utilizes magnetic bead-based technology can be processed manual or automated on open-ended liquid handling platforms as well as magnetic processors. MagNXT Cell free DNA Extraction Kit for professional, laboratory trained personnel use only.

Intended User

The assay is intended to be performed by a laboratory professional in clinical laboratory.

Test Principle

The MagNXT Cell free DNA Extraction Kit is based on superparamagnetic particle technology for the isolation of circulating cell-free DNA (cfDNA) from human plasma or serum. cfDNA, which serves as a biomarker for cancer, autoimmune diseases, and prenatal diagnostics, is selectively bound to magnetic particles under optimized buffer conditions. After a series of wash steps, purified cfDNA is eluted in a low-salt buffer compatible with downstream molecular applications such as PCR, qPCR, NGS, Southern blotting, and microarray analysis.

Summary

The MagNXT Cell-free DNA Extraction Kit is designed for the isolation of circulating cell-free DNA (cfDNA) from human plasma or serum samples using superparamagnetic particle-based technology. During the procedure, cfDNA binds selectively to magnetic particles under specific buffer conditions. Impurities and inhibitors are removed through a series of wash steps, and the purified cfDNA is then eluted in a low-salt buffer. The resulting high-quality cfDNA is compatible with a variety of downstream molecular biology applications, including PCR, qPCR, next-generation sequencing (NGS), Southern blotting, and microarray analysis. This extraction method enables for clinical and research purposes, including cancer monitoring, autoimmune disease evaluation, and prenatal diagnostics.

Materials Required But Not Provided

- Water bath or Heating block
- Micropipettes (Adjustable)
- Disposable barrier (Filter) pipette tips
- 1.5/2 ml microcentrifuge tubes (RNase and DNase free)
- Table top microcentrifuge
- Magnetic Separation Rack (can be supplied by Genes 2Me on request)
- Molecular biology grade ethanol (96-100%)
- Personal protective equipment (lab coat, goggles, etc)
- Disposable powder free gloves
- Vortexer

Storage, Operating Conditions and Stability

This product can be stored at room temperature (15-25 °C) for 18 months. If precipitate forms in any of the reagents of the kit, warm at 55 °C to dissolve. After reconstitution, Proteinase K needs to be stored at -20 °C.

* Store the MagPure particles at 2°C to 8°C after receiving the shipment.

Instruction Before Use

- Preheat a water bath or heating block to 55 °C.
- Add Protease Dissolve buffer to the Proteinase K, as shown on label, final concentration should be 20 mg/ml. For long term storage, the unused portion of the solution may be stored in aliquots at -20 °C until needed.
- Always vortex Proteinase K before use.
- Always vortex magnetic bead suspension before use.
- Dilute Buffer CW2 with an appropriate amount of ethanol (96-100 %) as shown on label and store at room temperature.

Sample Preparation Protocol

Collect ~1.5 ml blood sample in a streck tube and centrifuge it at 2500 xg for 10 min at 4 °C. After centrifugation, three different fractions are distinguished: the upper clear layer contains plasma; the intermediate buffy coat layer containing concentrated leukocytes, and the bottom layer containing concentrated erythrocytes.

Separate the plasma into a fresh microcentrifuge tube without disturbing buffy coat layer. Spin the microcentrifuge tubes at 16,000 xg for 10 min at 4°C. Discard the pellet and transfer the separated supernatant i.e., plasma into a fresh microcentrifuge tube.

MagNXT Cell free DNA Extraction Kit

Platform-Manual

SKU Code-G2M810003-50T, G2M810003-250T

Table 1. Kit Components

| Kit Contents | Kit Content Code | Kit Content Quantity | Kit Content Code | Kit Content Quantity |
|--------------------------|------------------|----------------------|------------------|----------------------|
| | 50 Tests | 50 Tests | 250 Tests | 250 Tests |
| MagPure Particles | G2MA3-3067-1 | 1 X 3 ml | G2MA3-3067-2 | 1 X 15 ml |
| Buffer CLB-3 | G2MA3-3068-1 | 1 X 65 ml | G2MA3-3068-2 | 1 X 315 ml |
| Buffer CW1 | G2MA3-3069-1 | 1 X 65 ml | G2MA3-3069-2 | 1 X 315 ml |
| Buffer CW2 | G2MA3-3070-1 | 2 X 10 ml | G2MA3-3070-2 | 1 X 75 ml |
| Buffer EB | G2MA3-3071-1 | 1 X 10 ml | G2MA3-3071-2 | 1 X 30 ml |
| Proteinase K | G2MA3-3072-1 | 1 X 60 mg | G2MA3-3072-2 | 1 X 300 mg |
| Carrier RNA | G2MA3-3073-1 | 1 X 310 µg | G2MA3-3073-2 | 1 X 310 µg |
| Protease Dissolve Buffer | G2MA3-3074-1 | 1 X 4 ml | G2MA3-3074-2 | 1 X 18 ml |

Manual Protocol

- 1). Transfer 400/500 µl of plasma into a fresh 1.5/2 ml microcentrifuge tube.
- 2). Add 700/900 µl Buffer CLB-3 along with 30/35 µl of Proteinase K (20 mg/ml) and 40/50 µl MagPure particle into the solution and mix thoroughly by inverting for 15-30 times. Mix upside down for 10 min at room temperature.
- * Optimal - Carrier RNA: Add 0.1-0.5 µg for each prep. Carrier RNA can reduce the adsorption of consumables on DNA, but carrier RNA may interfere with the DNA quantification using Qubit. Reducing the carrier RNA concentration to 100 ng will help in the accurate DNA quantification in Qubit.
- 3). Transfer the microcentrifuge tubes into a magnetic stand, and leave the tubes undisturbed for 3 min to adsorb the magnetic beads after short spin. Discard the supernatant.
4. Further add 800 µl of Buffer CW1 to the solution. Transfer to a magnetic stand and leave the tubes undisturbed for 2 min to attract magnetic beads after short spin. Discard the supernatant.
5. Further add 800 µl of Buffer CW2 to the solution. Transfer to a magnetic stand and leave the tubes undisturbed for 2 min for the magnetic beads to settle after discard the supernatant.
6. Repeat step 5.
7. Centrifuge, briefly, aspirate all solutions, and air dry for 5-10 min.
8. Add 30-50 µl Buffer EB and vortex to disperse the magnetic beads. Let the tubes undisturbed for 5-10 min, vortex several times to dissolve the nucleic acid.
9. Transfer the microcentrifuge tubes into a magnetic stand and let stand for 3 min after short spin.
10. Transfer the purified DNA into a new microcentrifuge tube and store DNA at -20°C or at -80°C for long time.

Troubleshooting Guide

A. Poor yield / low sensitivity

1) Incomplete sample lysis

- Sample mixed with Lysis Buffer and was not thoroughly homogenized. The mixture has to be shaken continuously. Alternatively, prolong the incubation time with Lysis Buffer.

2) Insufficient elution buffer volume

- Bead pellet must be covered completely with elution buffer and needs to be fully resuspended. Insufficient performance of elution buffer during elution step.
- Remove all buffers completely from the bead pellet after the binding and wash steps. Remaining buffer decreases the efficiency of the subsequent steps. Aspiration of adsorbed bead pellet.
- Do not disturb the attracted beads while aspirating the supernatant. This requires special caution when removing the lysate from the beads as the lysate is usually too opaque to allow visual control of the pellet.
- Time for magnetic separation too short or aspiration speed too high.

B. Low Purity/Low Sensitivity Insufficient washing procedure
















- Make sure that beads are resuspended completely during the washing procedure. If shaking is not sufficient to resuspend the beads completely mix by repeated pipetting up and down.

C. Poor performance of Nucleic acid in down-stream applications Carry-over of ethanol from wash buffers

- Be sure to remove all of the 80 % ethanolic wash solution from the final wash, as residual ethanol interferes with downstream applications. Ethanol evaporation from wash buffers.
- Close the buffer bottles tightly, avoid ethanol evaporation from buffer bottles as well as from buffer filled in reservoirs. Do not reuse buffer reservoirs.

Safety and Precautions

- **Chemical Handling:** Reagent cartridges/plates contain guanidine hydrochloride/guanidine thiocyanate, which may react with bleach to form highly reactive compounds. In case of spillage, clean with laboratory detergent and water.
- **Biological Samples:** Plasma/serum/blood may carry infectious diseases. Ensure all laboratory personnel are familiar with general safety guidelines for chemical usage, storage, and waste disposal. Refer to relevant Safety Data Sheets (SDS) for specific precautions.
- **Personal Protective Equipment (PPE):** Wear appropriate attire, including lab coats, gloves, goggles, and closed-toe shoes to protect against spills, splashes, and inhalation.
- **Ventilation:** Work in well-ventilated areas or use fume hoods to minimize exposure to harmful vapors or inhalation of chemicals. Handle the chemical waste in designated fume hoods.
- **Storage:** Store the chemicals correctly in designated areas, adhering to guidelines for temperature, compatibility, and segregation.
- **Labeling:** Ensure all the containers are clearly labeled with the chemical name, concentration, and hazard warnings to prevent accidents or confusion.
- **Handling:** Employ proper techniques, while handling chemicals, pour chemicals carefully and steadily to avoid spills, splashes, and excessive vapor release. Keep chemical containers closed while not in use.
- **Emergency Equipment:** Familiarize yourself with the location and proper use of safety showers, eyewash stations, fire extinguishers, and spill kits for swift response to emergencies.
- **Training:** All individuals must undergo comprehensive training on chemical handling, emergency procedures, and the use of safety equipment as per regulatory and institutional requirements before handling potentially biohazardous materials.

| Symbols for Use in the Labeling | |
|---|---|
| Symbols | Definition |
|  | KEEP AWAY FROM SUNLIGHT |
|  | TEMPERATURE LIMIT |
|  | IN VITRO DIAGNOSTIC MEDICAL DEVICE |
|  | UPWARD |
|  | CONSULT INSTRUCTIONS FOR USE |
|  | BATCH CODE |
|  | CATALOGUE NUMBER |
|  | USE BY DATE |
|  | DATE OF MANUFACTURE |
|  | MANUFACTURER |
|  | CONTAINS SUFFICIENT FOR <n> TESTS |
|  | CAUTION |
|  | DO NOT USE IF PACKAGE IS DAMAGED |
|  | AUTHORIZED REPRESENTATIVE IN THE EUROPEAN COMMUNITY/ EUROPEAN UNION |
|  | EUROPEAN CONFORMITY |



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