# VAMNE Magnetic Pathogen DNA Kit (Prepackaged)

**DM202**

Version 23.1

## Product Description

This kit is suitable for the isolation and purification of DNA from biological fluid samples (alveolar lavage fluid, blood, sputum, swab eluate, cerebrospinal fluid, etc.). It combines chemical and mechanical lysis methods and can efficiently lyse bacterial and fungal cells with thick cell walls. The kit uses high-affinity silica-coated magnetic beads, which adsorb nucleic acids in a high-salt buffer through hydrogen bonds and electrostatic forces. Unwanted proteins and salt ions are then rinsed away, and nucleic acids will be released in a low-salt elution buffer or Nuclease-free dd$H_2O$, enabling rapid isolation and purification of nucleic acids. The kit is compatible with an automatic nucleic acid extraction instrument (Vazyme #VNP-32P) that is based on the magnetic bead adsorption principle. Specially designed magnetic rods are used to adsorb, transfer, and release magnetic beads. By transferring the magnetic beads to the nucleic acid, the nucleic acid will be automatically extracted and purified. The DNA isolated with this kit is suitable for various downstream applications, including PCR, Real-Time PCR, metagenomic library preparation, and microarray analysis.

## Components

| Components | DM202-01 (64 T) |
|---|---|
| DNA Reagents (Prepackaged for DM202) | 4 × 16 T |
| Lysis Buffer 1 | 12.8 ml |
| Lysis Tube | 64 |
| Proteinase K | 3 × 1 ml |
| PBS | 12.8 ml |

## Storage

Proteinase K: Store at 2 ~ 8℃ and adjust the shipping method according to the destination.
Other components: Store at 15 ~ 25℃ and transport at room temperature.

## Applications

Blood samples: Fresh or frozen anticoagulated whole blood (≤ 200 µl);
Biological fluid samples: Fresh or frozen alveolar lavage fluid, sputum, cerebrospinal fluid, synovial fluid, pleural and peritoneal effusion, and vitreous humor (≤ $1 \times 10^7$ cells);
Swab samples: Fresh throat, nasal, and oral swab eluates (≤ $1 \times 10^7$ cells).

## Applicable Instruments

Automatic nucleic acid extraction instrument (Vazyme #VNP-32P) and other similar instruments (heating slots position: 1, 6, 7, 12).

## Self-prepared Materials

High speed centrifuge, vortex mixer, or homogenizer.

## Notes

For research use only. Not for use in diagnostic procedures.

1. Pathogen samples should be processed in a biosafety cabinet.
2. Disinfect the automatic nucleic acid extraction instrument with UV irradiation for at least 30 min before use (for nucleic acid extraction from pathogens, UV irradiation time may be extended to 12 h). After the experiment, wipe the inside of the extraction instrument with 75% ethanol and disinfect it with UV irradiation for 30 min.

## Experiment Process

**1. Preparation of prepackaged reagent**

Take out the prepackaged reagents from the kit, invert and mix several times to resuspend the magnetic beads. Gently shake the plate to make the reagents and magnetic beads sink to the bottom of the well. Please confirm the direction of the plate and be carefully tear off the aluminum foil sealing film.

▲ Avoid vibration when tearing off the sealing foil to prevent liquid from spilling.

**2. Genomic DNA extraction**

**2.1 Pathogen genomic DNA extraction**

**Sample pretreatment:**

a. Blood samples: Add 200 µl of blood sample to a Lysis Tube.

▲ Add to 200 µl with PBS if the sample volume is less than 200 µl.

b. Sputum samples: Add 5 volumes of N-acetylcysteine solution (10 g/L) (not provided) to an appropriate amount of sputum and vortex for 30 - 60 min to liquefy. Centrifuge at 7,500 rpm (5,400 × g) for 10 min and discard the supernatant. Resuspend the pellet in 200 µl of PBS and transfer the mixture to a Lysis Tube.

c. Swab and other biological fluid samples: Add 200 µl of sample to a Lysis Tube.

▲ Add to 200 µl with PBS if the sample volume is less than 200 µl.

▲ If the sample volume is more than 200 µl, transfer the sample to a 1.5 ml Nuclease-free centrifuge tube. Centrifuge at 12,000 rpm (13,800 × g) for 5 min and discard the supernatant. Resuspend the pellet in 200 µl of PBS and transfer the mixture to a Lysis Tube.

**Sample extraction:**

Add 200 µl of Lysis Buffer 1 and then 40 µl of Proteinase K to the above Lysis Tube containing the sample. Vortex the Lysis Tube on a mixer at maximum speed for 10 min. After vortexing, centrifuge at 12,000 rpm (13,800 × g) for 3 min. Transfer all the supernatant to the wells in Columns 1 or 7 of the 96 deep well plate, and proceed to Step 3.

▲If there is still foam after centrifugation at 12,000 rpm (13,800 × g) for 3 min, increase the centrifugation time to remove the foam.

**2.2 Blood sample total genomic DNA extraction**

Add 200 µl of the blood sample and then 40 µl of Proteinase K to the wells in Columns 1 or 7 of the 96 deep well plate, and proceed to Step 3.

**3. Operation of the automatic instrument**

a. Place the 96 deep well plate into the nucleic acid extraction instrument in the correct orientation (with the notch facing the upper left). Put on the magnetic bar sleeves, and ensure them fully envelops the magnetic rods.

b. Set the program as follows (or select the corresponding preset) for automatic extraction:

| Step | Slot Position | Name | Mixing Time (min) | Adsorption Time (sec) | Waiting Time (min) | Volume (µl) | Mixing Speed | Temperature (℃) | Mixing Position | Mixing Amplitude | Adsorption Position | Adsorption Speed |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 2 | Moving magnetic beads | 1 | 60 | 0 | 700 | 8 | - | 10% | 80% | 0% | 5 |
| 2 | 1 | Lysis | 10 | 60 | 0 | 790/990 | 10 | 70 | 10% | 100% | 0% | 5 |
| 3 | 3 | Washing 1 | 3 | 60 | 0 | 700 | 10 | - | 10% | 100% | 0% | 10 |
| 4 | 4 | Washing 2 | 1 | 60 | 0 | 700 | 10 | - | 10% | 100% | 0% | 10 |
| 5 | 5 | Washing 2 | 1 | 60 | 1 | 700 | 10 | - | 10% | 100% | 0% | 10 |
| 6 | 6 | Elute | 15 | 220 | 0 | 110 | 10 | 70 | 10% | 100% | 0% | 10 |
| 7 | 2 | Discarding magnetic beads | 0.1 | 0 | 0 | 700 | 8 | - | 10% | 80% | 0% | 5 |
| Other settings (in the Option menu): Heating settings (heating and action start at the same time); Adsorption settings (three-stage adsorption) | | | | | | | | | | | | |

c. At the end of the automated procedure, transfer the eluent in Columns 6 or 12 to clean Nuclease-free centrifuge tubes for direct use in downstream experiments or storage at -20℃.