Nucleic Acid Extraction Kit (Magnetic Bead Method)

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Language	English
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Applicable Instruments	Zybio EXM3000/3200, Zybio EXM6000/6200, TIANLONG NP968-C
Specimen Types	Serum, plasma, nasopharyngeal swab, cell preservation fluid, tissue fluid, urine and secretions
Reagent Storage	2~8°C
Shelf-life Stability	12 months

Intended Use

The kit is intended for extraction and purification of viral nucleic acids (DNA/RNA) from serum, plasma, nasopharyngeal swab, cell preservation fluid, tissue fluid, urine and secretions specimens using magnetic beads.

Test Principles

The surface of the beads in the kit has specific polymeric groups that can capable of absorbing nucleic acid (DNA/RNA). In special conditions such as hypersaline, cells or viruses in the sample rapidly lyse and release their nucleic acids. Then the nucleic acids are adsorbed specifically by the magnetic beads. The nucleic acids on the beads will be separated from the liquid phase by using a magnetic stand. The beads are washed by using Extraction Reagent II to remove impurities and inhibitors in the liquid phase. Finally, the nucleic acid is eluted from the beads by changing the liquid phase conditions, so as to achieve rapid and efficient separation of the nucleic acids.

Materials Provided

REF	Specification	Packing Size	Applicable Instruments
01.09.20.00.B2.17	B-200	8 T/Kit	Zybio EXM3000/3200, TIANLONG NP968-C
01.09.20.00.B2.10	B-200	16 T/Kit	
01.09.20.00.B2.12	B-200	20 T/Kit	
01.09.20.00.B2.08	B-200	32 T/Kit	
01.09.20.00.T1.09	T-200	32 T/Kit	Zybio EXM6000/6200
01.09.20.00.T1.07	T-200	96 T/Kit	

Main Components

Table 1 Main Components of B-200

Constituents	B-200				Main Components
	8 T/Kit	16 T/Kit	20 T/Kit	32 T/Kit	
Proteinase K	120 μL× 1 tube	240 μL× 1 tube	300 μL× 1 tube	480 μL× 1 tube	15mg/mL-35mg/mL Proteinase K
Nucleic Acid Extraction Reagent Prepackaged in 96-Well Plates	4T× 2 plates	8T× 2 plates	1T× 20 plates	16T× 2 plates	Extraction Reagent I, Extraction Reagent II, Elution Buffer, Magnetic Beads Solution
8-strip magnetic tips comb	4 pcs	4 pcs	10 pcs	4 pcs	/

Table 2 Main Components of T-200

Constituents	T-200		Main Components
	32 T/Kit	96T/Kit	
Extraction Reagent I	32T × 1 plate	96T × 1 plate	15%-35% Guanidine thiocyanate, 45%-55% Isopropyl alcohol
Extraction Reagent II	32T × 1 plate	96T × 1 plate	65mM-75mM Tris buffer, 600mM-800mM Sodium chloride
Elution Buffer	32T × 1 plate	96T × 1 plate	Deionized water
Magnetic Beads Solution	32T × 1 plate	96T × 1 plate	0.08%-0.4% Magnetic beads
Proteinase K	480μL× 1 tube	1440μL× 1 tube	15mg/mL-35mg/mL Proteinase K

Warnings and Precautions

- For in vitro diagnostic use only.
- Please read instructions for use carefully before use, and it is for professional use only.
- The 96-well plate is for single use. [Proteinase K] is reusable.
- Protective equipment accessories (goggles, work clothes, hats, shoes, gloves, etc.) should be worn during operation.
- Use sterile centrifuge tubes and filter-tips for sample preparation.
- All specimen to be tested should be considered as infectious substances and processed strictly in accordance with laboratory biosafety requirements. Repeated freezing and thawing specimens should be avoided, mixed well before use.
- The disposal of waste liquid should be in accordance with local laws and regulation.
- The components need to be mixed well before use. In case of precipitation in [Extraction Reagent I], it can be used after sufficient dissolution at room temperature.
- ⚠ [Extraction Reagent I] contains guanidine thiocyanate. Please take care to protect yourself from skin or eye damage when using.
- [Extraction Reagent I] contains isopropyl alcohol. Please keep away from fire sources or other risk factors.
- Please do not place the kit below -20 °C. If this situation cannot be avoided, it should be ensured that the freeze-thaw of kit no more than 5 times.
- After the operation, the work area surface and the instrument surface should be disinfected with a freshly prepared 10% sodium hypochlorite solution, and then cleaned with 75% ethanol or pure water. Finally, turn on UV light (if available) to disinfect working surfaces for 30 minutes.
- Check the integrity of the reagent before use. If the 96-well plate is found to have leakage, insufficient volume, no reagents in the wells, etc., please stop using such kit. Use a new kit for extraction.
- 96-well plates of B-200 specifications and 96-well plates of T-200 specification [Extraction Reagent II] may have sealing film difficult to remove, partial film residual on the plate, plate deformation, etc., please stop using such kit. Use a new kit for extraction.
- Any serious incident that has occurred when using shall be reported to the manufacturer or the competent authority of the Member State in which the user is established.

Nucleic Acid Extraction Kit (Magnetic Bead Method)

Storage & Stability

- The kit can be stored at 2~8°C for 12 months.
- Once opened, 96-Well plates are disposable.

Sample Requirements

- Samples should be collected, transported, stored, and processed according to the relevant regulatory requirements.
- Sample types: Liquid samples such as serum, plasma, nasopharyngeal swab, cell preservation fluid, tissue fluid, urine and secretions.
- The collected samples can be used for nucleic acid extraction immediately, or stored at 2~8°C for no more than 24 hours, stored at -70°C or below for a long term. Repeated freezing and thawing specimens should be avoided. Frozen samples need to be thawed and mixed well before use.
- The samples should be transported in sealed cooler box or styrofoam box with ice seal.

Sample Preparation

This kit requires 200 μL of sample for a single determination. Place samples in a biological safety cabinet. If the sample is frozen, completely thaw it at room temperature before use.

Extraction Procedure

1. Extraction Procedure with EXM3000/3200, TIANLONG NP968-C (For B-200)

   Please follow EXM3000/3200 Nucleic Acid Isolation System User Manual, Nucleic Acid Extractor NP968-C User Manual for extraction setup.

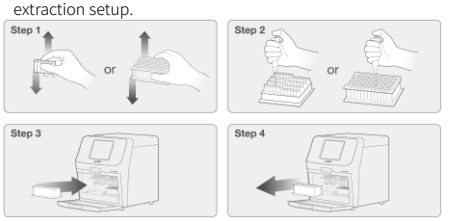

Figure 1 Operation Method of B-200

1) Take out all the components in the kit. Mix the 96-well plate upside down so that the liquid adhering to the sealing film and the wall of the wells of the 96-well plate falls to the bottom of the plate. Let them stand for 3-5 minutes for use.
2) Carefully open the sealing film of the 96-well plate, and add 15 μL [Proteinase K] to the position A1~H1 and A7~H7 sequentially, then add 200 μL sample sequentially. Place the 96-well plate in the corresponding position of the instrument and insert the 8-strip magnetic tips comb. Close instrument door after finish loading.

   Exception: *For B-200-20, label facing the operator, carefully open the sealing film of the 6-well reagent cards, and add 15 μL [Proteinase K] and 200 μL sample to the position 1. Then put the reagent cards into the baseplate crosswise (as shown in Figure 2), put the baseplate into the corresponding position of the instrument, and insert 8-strip magnetic tips comb. Close instrument door after finish loading. The next operations are the same.*
3) Turn on the nucleic acid Extraction system, enter the page < Program Edit >, and set the extraction process according to table 3:

Table 3 Running Program Setting

No.	Position	Name	Waiting Time (min)	Mixing Time (min)	Absorption Magnetic Beads Time (sec)	Mixture Velocity	Volume	Temperature (℃)
1	2	Move	0	0	30	Slow	150	0
2	1	Lysis	0	4	60	Medium	500	65
3	3	Wash	0	1	60	Medium	600	0
4	6	Elution	0	2	30	Medium	50	85
5	1	Move	0	0	0	Slow	150	0

Note: *The parameters of volume set according to the actual extraction effect, may not be the same with reagent volumes. Please carefully check that the 8-strip magnetic tips comb has been inserted into the appropriate position before the program runs.*

For EXM3000/3200 Nucleic Acid Isolation System: Turn on the nucleic acid Extraction system, select the <ZYBIO-B-200> or <ZYBIO-B-200Q> program. (<ZYBIO-B-200> is the standard program and <ZYBIO-B-200Q> is the quick program. Choose the appropriate program according to the requirements.)

4) Click "Start" to run the extraction program. The process takes about 9-12 minutes.
5) Transfer the nucleic acid solution at the A6-H6 and A12-H12 positions of the 96-well plate to 1.5 mL centrifuge tube for further use (a small amount of beads can be removed by centrifuge or magnetic stand).

2. Extraction Procedure with EXM6000/6200 (For T-200)

   Please follow EXM6000/6200 Nucleic Acid Isolation System User Manual for extraction setup.

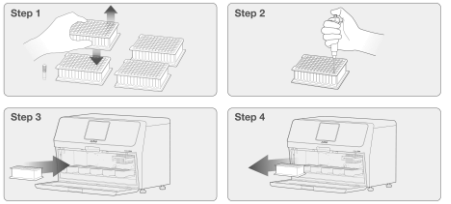

Figure 2 Operation Method of T-200

1) Take out all the components in the kit. Mix the 96-well plate upside down so that the liquid adhering to the sealing film and the wall of the wells of the 96-well plate falls to the bottom of the plate. Let them stand for 3-5 minutes for use.
2) Carefully open the sealing film of the 96-well plate. According the number of the samples, add 15 μL [Proteinase K] sequentially to the 96-well plate of [Extraction Reagent I], then add 200 μL sample sequentially.
3) Put the 96-well plate of [Extraction Reagent I] into the NO.1 position of the instrument. Put the 96-well plate of [Magnetic Beads Solution] into the NO.2 position of the instrument and steadily insert the 96-strip magnetic tips comb. Put the 96-well plate of [Extraction Reagent II] into the NO.3 position of the instrument. Put the 96-well plate of [Elution Buffer] into the NO.5 position of the instrument. The NO.4 position is empty. Close instrument door after finish loading.

   Note: *Please carefully check that the 96-strip magnetic tips comb has been inserted into the appropriate position before the program runs.*
4) Turn on the nucleic acid extraction system, select the <ZYBIO-T-200> program.

5) Click "Start" to run the extraction program. The process takes about 12 minutes.
6) Take out the 96-well plate at the NO.5 position of the instrument, and the nucleic acid solution can be used directly for further use, or transferred to 1.5 mL centrifuge tube for use (a small amount of beads can be removed by centrifuge or magnetic stand).

Limitations

This product needs to be used with a magnetic stand in manual operation.

Performance Characteristics

- **Extraction efficiency:** Samples containing ≥10 IU/mL DNA or ≥15 IU/mL RNA viruses can be extracted. The extracted products can be directly used for subsequent molecular biology experiments such as PCR, hybridization, and sequencing.
- **Precision:** Samples containing ≥200 IU/mL DNA or ≥500 IU/mL RNA virus were extracted with a Ct value coefficient of variation CV≤5%.

References

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2. Sonja B. Magnetic particles for the separation and purification of nucleic acids. Appl Microbiol Biotechnol, 2006, 73:495-504.
3. Li J M. Real-time Fluorescent PCR Technology [M]. Beijing: People's Military Medical Press, 2007.

Symbol Interpretation

Symbol	Title and Description	Symbol	Title and Description
IVD	In vitro diagnostic medical device	LOT	Batch code
	Consult instructions for use		Use-by date
CE	CE marking of conformity		Manufacturer
EC REP	Authorized Representative in the European Community		Temperature limit
REF	Catalogue number		Warning
	Date of Manufacture		

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CE IVD
Nucleic Acid Extraction Kit (Magnetic Bead Method)

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