Jiangsu Bioperfectus Technologies Co., Ltd.

Pathogen DNA/RNA Extraction Kit (Magnetic Bead Method) INSTRUCTIONS FOR USE

For Research Use Only (RUO)
For use on the SSNP-2000A, SSNP-2000B, SSNP-3000A, SSNP-9600A, SMPE-960, SAW-48, SAW-96, SHT-Auto960 and other automatic nucleic acid extraction systems manufactured by Jiangsu Bioperfectus Technologies Co., Ltd.

REF SDK60120-48T Σ 48

1. Intended Use

This product is suitable for the extraction and purification of total pathogen nucleic acid from samples such as sputum, whole blood, serum/plasma, nasal / pharyngeal /oral swab, urine sediment, tissue homogenate supernatant, etc. The purified DNA/RNA can be used for clinical in vitro detection.

2. Kit Components

Name	48 T/ kit		Reagent and Fill Volume			
96-deep-well pre-packed extraction reagents	8 T/plate × 6	16 T/plate × 3	**Column**	**Solution name**	**Components**	**Volume**
			Column 1/7	Lysis binding buffer	Guanidine isothiocyanate	600μL
			Column 3/9	Wash buffer 1	Guanidine hydrochloride	750μL
			Column 4/10	Wash buffer 2	Ethanol	750μL
			Column 5/11	Magnetic bead buffer	Magnetic beads	400μL
			Column 6/12	Elution buffer	RNase-Free water	75μL
Proteinase K	1 vial	1 vial	1 mL/vial			
Bead tube	48 tubes	48 tubes	0.5g/tube			
Magnetic rod stirring sleeve	6 strips	6 strips	Eight magnetic rod stirring sleeve			

3. Storage and stability

The kit can be transported at -20°C~45°C and stored at 4°C~30°C away from light, with a shelf life of 18 months. See the product label for the production date and expiration date.

4. Materials and devices required but not provided

Appropriate automatic nucleic acid extraction systems manufactured by Jiangsu Bioperfectus Technologies Co., Ltd., such as SSNP-2000A, SSNP-2000B, SSNP-3000A, SSNP-9600A, SMPE-960, SAW-48, SAW-96, SHT-Auto960, etc.

5. Background Information

The magnetic bead method of nucleic acid extraction has advantages that cannot be matched by traditional DNA extraction methods. It is mainly embodied in: ①It can realize automation and large-volume operations. At present, there are automatic nucleic acid extraction instruments, which can achieve multiple samples with one sample extraction time. The processing meets the biological high-throughput operation requirements, enabling rapid and timely response to infectious disease outbreaks. This feature makes traditional methods unmatched; ②Simpler operation and shorter time, the entire extraction process has only four steps; ③Safe and non-toxic, without the use of toxic reagents such as benzene and chloroform in traditional methods, the hazard to experimental operators is reduced to a minimum, fully in line with modern environmental protection concepts; ④Magnetic beads specifically bind to nucleic acid, making the extracted nucleic acid with high purity and concentration.

6. Technical Principle

This kit uses magnetic beads and a buffer system with a unique isolation function and is used in conjunction with a nucleic acid extraction system to isolate and purify high-quality nucleic acids from samples. The particularly-coated magnetic beads have a strong affinity to nucleic acids in the sample under certain conditions. When these conditions change, the magnetic beads release the nucleic acids absorbed by them to extract and purify nucleic acids quickly.
This product is based on the purification method of glass beads grinding and proteinase K decomposition to efficiently lysate bacterial and fungal cells with thick cell walls, and realize DNA/RNA co-extraction of bacteria, fungi, viruses, mycoplasma and other pathogenic microorganisms. It is suitable for downstream PCR, qPCR, library preparation and other applications.

7. Precautions and disposal requirements

7.1 Warnings and Precautions

- Please read the IFU carefully before operation, and operate strictly in accordance with the IFU.
- Please use the unpacked reagents promptly to prevent reagents from volatilization and ensure extraction performance.
- There may be residual magnetic beads in the columns 6/12. When aspirating nucleic acid for downstream experiments, it is advisable to avoid inhaling magnetic beads as much as possible.
- Avoid shaking the reagent violently to prevent excessive foam.
- The guanidine salts in the kit are corrosive and irritating. In the case of accidental skin exposure, immediately rinse with plenty of water, or go to the hospital if it is serious.
- Kits of different lots cannot be used interchangeably, and all kits should be used within their shelf life.
- The operators should be trained and qualified for molecular biological testing. The laboratory should be equipped with proper biosafety protection facilities and procedures.

7.2 Disposal

- The product and specimens should be considered infectious and disposed in accordance with local regulations.
- Packaging materials that are non-degradable should be collected and delivered to the local process center.

7.3 Vigilance

If any serious incident has occurred in relation to this product, please contact the manufacturer and report to the local competent authority.

8. Sample requirements

- This kit is suitable for the extraction and purification of total pathogen nucleic acid from samples such as sputum, whole blood, serum/plasma, nasal / pharyngeal /oral swab, urine sediment, tissue homogenate supernatant, etc.
- Fully mix the sample before extraction to prevent the sample's heterogeneity from affecting the amounts of nucleic acids extracted.
- For use with other samples, please contact our technical service department for relevant information.

9. Extraction Procedure

1) Sample pretreatment:
a) Sputum and other sticky samples should be liquefied with DTT solution (0.1%) or N-acetylcysteine solution (10 g/L) (reagents are not provided). NaOH liquefaction is not recommended for the extraction of total pathogen nucleic acid. The samples after liquefaction should be enriched: absorb 1-2 mL of the samples after liquefaction, centrifuge at 10,000 rpm for 3 min and discard part of the supernatant, retain 500-600 μL and resuspend the precipitate for later use.
Note: If only the DNA of pathogens with thick cell walls such as *Mycobacterium tuberculosis* is extracted, it can be liquefied with 4% NaOH, and after centrifugation, discard the supernatant and resuspend the precipitate in 500-600 μL normal saline or PBS.
b) Blood extracellular viruses are recommended using serum/plasma samples, which can skip the pretreatment step and go directly to Step 5 for extraction. If the sample is whole blood, it is recommended to centrifuge, extract the upper plasma and middle white blood cell layer. The absorption of small amounts of red blood cells does not affect the subsequent extraction.
c) Samples with a small number of cells should be centrifuged and enriched first: add 1-2 mL samples into a centrifuge tube, centrifuge at 10,000 rpm for 3 min, and discard part of the supernatant, retain 500-600 μL and resuspend the precipitate for later use.
2) Add 500 μL sample (sputum liquefaction solution, biological fluids, swab eluate, culture solution, etc.) and 20μL Proteinase K to the Bead tube;
3) Use a vortex mixer at maximum speed for 10 min or a grinder to grind for 90 s, and briefly centrifuge;
4) (Optional) Incubate at 55°C for 10 min, briefly centrifuge to collect the droplets on the tube;
Note: If there is still turbid after centrifugation, please centrifuge at 10,000 rpm for 3 min again, and then extract the supernatant.
5) Take out the reagent plate, and peel off the aluminum seal. Add 300-400 μL supernatant to the corresponding wells in columns 1 or 7 of the reagent plate;
6) Load the plate into the nucleic acid extraction system with the notch of the plate facing outward;
7) Insert magnetic rod stirring sleeves;
8) Close the test chamber;
9) The steps are shown in the following table, click Run;
10) After the test, transfer the extracted nucleic acids in columns 6 and 12 to clean Nuclease-free centrifuge tubes.

Step	Name	Well	Adsorption time (s)	Mixing time (s)	Mixing speed	Volume
1	/	1/7	0	0	1	30μL
2	Magnetic beads resuspending	5/11	0	60	25	400μL
3	Binding	1/7	12	540	25	900μL
4	Wash 1	3/9	12	60	25	750μL
5	Wash 2	4/10	12	60	25	750μL
6	/	4/10	0	0	1	30μL
7	Elution	6/12	12	180	25	75μL
8	Abandoned magnetic beads	4/10	30	60	25	750μL
Associated Settings		Lysis temperature: 25°C		Elution temperature: 85°C	Dry time: 180s	

10. Performance Characteristics

The CV value of extraction precision of the same batch or different batches is less than 5%.

11. Symbols

The following symbols are used in labeling for the products.

LOT	Batch code	REF	Catalogue number
	This side up	Σ	Contains sufficient for <n> tests

	Manufacturer		Consult instructions for use
	Date of manufacture		Use-by date
	Temperature limit		Keep away from sunlight
	Do not reuse		

12. Technical Support

For detailed programming instructions regarding the use of the kit please contact our Technical Support at E-mail: support @bioperfectus.com.

13. References

[1] Levison P R, Badger S E, Dennis J, et al. Recent developments of magnetic beads for use in nucleic acid purification [J]. J Chromatogr A, 1998, 816:107-111.

[2] Zhang H P, Bai S, Xu L, et al. Fabrication of mono-sized magnetic anion exchange beads for plasmid DNA purification [J]. J Chromatogr B Analyt Technol Biomed Life Sci. 2009, 877(3):127-133.

14. Revision

Revision MM/YYYY	**Change description**
A 11/2023	First Publishing.