Mag Beads Bacterial Genomic DNA Extraction Kit

【Product Name】

Mag Beads Bacterial Genomic DNA Extraction Kit

【Packaging Specifications】

20T/box（Art.No.GP611-20）；100T/box（Art.No.GP611-100）；

300T/box（Art.No.GP611-300）

【Intended Use】

This reagent kit is designed for efficient extraction of genomic DNA from Gram-negative bacteria and Gram-positive bacteria. It is suitable for use in both automated and semi-automated nucleic acid extraction workstations. The extracted DNA can be utilized in various downstream molecular biology experiments, including PCR, Southern blot analysis, fluorescence quantitative PCR, and other applications.

【Detection Principle】

DNA binds to the surface of Magbeads coated with silicon under high salt conditions. After multiple washes to remove impurities such as proteins, DNA is eluted under low salt conditions, resulting in high-purity genomic DNA.

【Main Components】

Component	GP611-20	GP611-50	GP611-100	GP611-300
Suspension	2.5mL	6mL	12.5mL	37mL
Lysate	9mL	21mL	45mL	135mL
Washing buffer	6mL(+7.8mL Anhydrous ethanol)	15mL(+19.5mL Anhydrous ethanol)	30mL(+39mL Anhydrous ethanol)	90mL(+117mL Anhydrous ethanol)
Eluant	3mL	7mL	15mL	45mL
Beads	450μL	1.05mL	2*1.05mL	6.5mL
Proteinase K	450μL	1.05mL	2*1.05mL	6.5mL
RNase A	120μL	300μL	600μL	1.8mL

【Storage Conditions and Shelf Life】

Magbeads suspension should be stored at 2-8°C, while Proteinase K and RNase A should be stored below 4°C. The remaining reagents can be stored at room temperature, and the shelf life for all components is 1 year;

Transportation is permitted between 4-37°C, with a maximum transit time of 14 days.

【Required Equipment and Reagents】

1. Common equipment: Centrifuge, pipettor, vortex mixer, shaking incubator (or a constant temperature water bath), centrifuge tubes, etc.;
2. Equipment for automated extraction: Nucleic acid extractor (or other fully automated nucleic acid extractors) with matching 96-well deep plates and magnetic rod covers;
3. Equipment for manual extraction: 16-tube magnetic rack (for sample counts ≤ 16);
4. Reagents: 80% ethanol, lysozyme, isopropanol.

【Precautions】

1. Carefully read the instructions for this reagent kit before conducting the experiment and strictly follow the instructions.
2. Do not freeze the magnetic beads, and thoroughly mix the magnetic bead suspension before use.
3. Before each use, check for any precipitation in the components. If present, redissolve at 37°C.
4. Before initial use, add quantified anhydrous ethanol according to the label instructions for the washing solution.

【Manual Single-Tube Operation Steps】

1. Sample Lysis:

1) For Gram-negative bacteria: Take 1-5mL bacterial culture, centrifuge at 10,000 rpm for 1 min, and aspirate the supernatant as much as possible.

For Gram-positive bacteria: Take 1-5mL bacterial culture, centrifuge at 10,000 rpm for 1 min, aspirate the supernatant as much as possible, add 100μL suspension and 15μL lysozyme, incubate at 37°C for 1h, mixing 3-5 times during the incubation.

2) Add 5μL RNase A, 400μL lysis buffer, and 20μL Proteinase K solution to the bacterial pellet or the bacterial suspension after incubation, vortex and shake well. Incubate at 60°C for 15 minutes, vortex-mixing every 3 minutes during incubation (or incubate at 60°C with constant shaking for 15 minutes).

2. DNA Binding: Add 300μL isopropanol and 20μL magnetic bead suspension, shake well at room temperature for 10 minutes.

3. Magnetic Separation: Place the centrifuge tube on the magnetic rack, invert 2-3 times, let the magnetic beads fully adsorb, completely remove the supernatant (keep the centrifuge tube fixed on the magnetic rack throughout the process), avoid contact with the magnetic beads.

4. Wash with Washing Solution: Remove the centrifuge tube from the magnetic rack, add 600μL washing solution, cover the centrifuge tube cap, vortex-mix for 10s to ensure thorough mixing of the magnetic beads, then vortex-mix for 1-2 minutes for magnetic separation.

5. Wash with 80% Ethanol: Wash twice using 600μL 80% ethanol with the same procedure as above.

6. Ethanol Removal: Place the centrifuge tube on the magnetic rack, let it stand for 5-10 minutes in a ventilated and dry place.

7. Elution: Remove the centrifuge tube from the magnetic rack, add 50-100μL elution buffer, vortex-mix for 20s, ensuring thorough mixing of the magnetic beads with the elution buffer. Place the centrifuge tube in a 60°C water bath for 10 minutes, vortex-mixing four times during this period (or incubate at 60°C with constant shaking for 10 minutes).

8. Nucleic Acid Transfer: Place the centrifuge tube on the magnetic rack and let it stand for 1 minute. After the magnetic beads are fully adsorbed, transfer the supernatant to a new centrifuge tube and store at -20°C for later use.

【Automated 16/32-Channel Nucleic Acid Extractor Operating Procedure】

1. Sample Preparation: In a 96-well plate, add the specified amounts for each corresponding well according to the table below, simultaneously processing 16/32 samples.

Position	1、7	2、8	3、9	4、10	5、11	6、12
Reagent	Isopropanol（300μL）	Beads（20μL）、Water（280μL）	Washing buffer（600μL）	80% Ethanol（600μL）		Eluant（50-100μL）

2. Sample processing: refers to the steps in the "Single-Tube Manual Operation":

3. Automated Extraction: Transfer the processed samples to the 1st and 7th columns of a 96-well plate. Then, place the prepared 96-well sample plate into the QP-AUT-32 nucleic acid extractor, insert the magnetic rod sleeve, open the instrument's operating program, select the corresponding program, click "Run," and start the extraction process.

4. Nucleic Acid Transfer: After the instrument operation is complete, transfer the eluate from position 6 (or 12) to a clean centrifuge tube free of nucleases.

The parameters for the 32-channel nucleic acid extractor (QP-AUT-32) program are set as follows

Step	Site	Name	Waiting time(min)	Mixing time（min）	Suction time(sec)	Volume (μL)	Mixing velocity	Ttemperature
1	1	Transfer beads	0	0	60	300	3	OFF
2	2	Comebine DNA	0	10	40	800	3	OFF
3	3	Washing 1	0	2	40	600	3	OFF
4	4	80% Ethanol	0	1	40	600	3	OFF
5	5	80% Ethanol	0	1	40	600	3	OFF
6	6	Elution	3	10	60	100	3	60℃
7	4	Abandon beads	0	1	0	600	3	OFF

【Automated 96-Channel Nucleic Acid Extractor Operating Procedure】

1. **Sample Preparation:** In a 96-well plate, add the specified amounts for each corresponding well according to the table below, simultaneously processing 96 samples.

Sosition	1	2	3	4	5	6
Reagent	Absolute ethyl alcohol （300μL）	Beads（20μL）、Water（280μL）	Washing buffer 1 （600μL）	Washing buffer 2 （600μL）	80% Ethanol （600μL）	Eluant （50-100μL）

2. Sample Processing

Refer to the steps in the "Single-Tube Manual Operation" for this stage.

3. Automated Extraction

Transfer the lysate mixture to a 96-well deep plate at position 1. Then, sequentially place the prepared 96-well sample plate into the QN-AUT-96 nucleic acid extractor or a similar type of extractor, insert the magnetic rod sleeve, open the instrument's operating program, select the corresponding program, click "Run," and start the extraction process.

4. Nucleic Acid Transfer

After the instrument operation is complete, directly seal or transfer the eluate from position 6 to a clean centrifuge tube free of nucleases, and store at -20°C for future use.

The parameters for the 96-channel nucleic acid extractor (QP-AUT-96) program are set as follows

Procedure	Step 1	Step 2	Step 3	Step 4	Step 5	Step 6	Step 7
Station	2	1	3	4	5	6	5
Waiting time	00:00:00	00:00:00	00:00:00	00:00:00	00:00:00	00:03:00	00:00:00
Mixed model	3	3	3	3	3	3	3
Mixing time	00:00:00	00:10:00	00:02:00	00:01:00	00:01:00	00:10:00	00:00:30
Suspend	No	No	No	No	No	No	No
Suction time	00:01:00	00:01:00	00:01:00	00:01:00	00:01:00	00:01:00	00:00:00
Volume	300μL	1000 μL	600 μL	600 μL	600 μL	100 μL	600 μL
Temperature	—	—	—	—	—	60℃	—

【Basic Information】

Version Number: 1.1

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