

Bacterial Genomic DNA Extraction Kit (Magnetic Bead Method)

Cat No.: DM1600

Package: 50T/100T

Storage: 2-8°C (Note: RNase A is stored in accessory form, -20°C; proteinase K, magnetic beads, 4°C storage, do not move cold)

not move cold)			VNV 28°	
Component	50T	100T C	Storage	
Lysate	10 mL	20 mL	RT	
Binding Buffer	15 mL	30 mL	RT	
Wash Buffer1	30 mL	60 mL	RT	
Wash Buffer2	7.5 mL	0 15 mL	RT	
Proteinase K	20mg×1	20mg×2	2-8°C	
Lysozyme Liquid	1mL×1	1mL×2	RT	
RNase A	100uL	100uL×2	-20°C	
Elution Buffer	10mL	10mL	RT	
Magnetic Bead	1mL×1	1mL×2	2-8°C, Do not freeze	
Instruction	1	1	-	

Product description:

Magnetic bead method bacterial genome DNA cassette, make bacteria in the lysate lysis completely, under the action of binding liquid to promote genomic DNA and magnetic beads specifically identify and efficient binding, after the rinse can maximize remove impurity protein and other organic compounds, under the action of external magnetic field force can separate DNA from the sample. The extracted genomic DNA with high purity A260/ A280 ratio between 1.8 and 2.0 can be applied to downstream experiments of various types of molecular biology. Suitable for Gram-negative bacteria.

Advantages:

The extraction of bacterial genomic DNA by magnetic bead method has incomparable advantages to the traditional column method. Compared with the column method, it greatly reduces the experimental time and extracts high-quality bacterial genomic DNA that can be applied to all kinds of molecular biology downstream experiments. It has the advantages of simple operation, short time, safe and non-toxic, can complete automatic extraction and so on.

Operation steps (for reference only):

Before use, add isopropanalcohol / absolute ethanol to the rinse solution, refer to the label on the bottle (50T / 100T rinse 1 with 30 mL / 60 mL isopropanalcohol alone, 50T / 100T rinse 2 with 22.5mL/45mL absolute ethanol alone).

Protease K solution configuration: add 1 mL lyase solution into 20mg protease K powder, mix 20 mg/mL protease K solution, and keep the protease solution-20°C.

1. 1-3mL of bacterial solution was added to the EP tube, centrifuged at 12000rpm for 1min, and the supernatant was aspirated as far as possible.

2. Add 200μ L of lysate to the bacteria, vortex and shake, or blow the straw body repeatedly with a pipete gun to fully suspend the bacteria. Add 20μ L proteinase K, fully mix well, digest in 60°C water bath for 10-20min, the

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bacterial solution is clear and thick, which is completely digested.

3. The EP tube was removed from the water bath and the temperature dropped to room temperature for $2\mu L$ RNase A, mixing the pipette gun and let at room temperature for 15min.

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4. Add an equal volume of binding liquid, blow and mix well with a pipette gun, and take a water bath in a 60°C water bath for 10min.

5. The EP tube was removed from the water bath, and 20μ L magnetic beads were added after dropping to room temperature, vortex and shaken at room temperature for 10min. Place the EP tube in the magnetic frame for magnetic separation. After the magnetic beads are fully adsorbed to the magnetic frame, remove the liquid along the tube wall with a pipette gun. Be careful not to absorb the magnetic beads.

6 Add 500μ L of rinse solution 1 (isopropyl alcohol before use) and mix well with a vortex oscillator. Place the centrifuge tube in the magnetic frame. After the magnetic beads are completely adsorbed to the magnetic frame, remove the liquid along the pipe wall with a pipete gun. Be careful not to absorb the magnetic beads.

7、 Repeat step 6.

8 Add 500µL of rinse sing 2 (with absolute ethanol) and mix with a vortex oscillator. Place the centrifuge tube in the magnetic frame. After the magnetic beads are completely adsorbed to the magnetic frame, remove the liquid along the tube wall with a pipete gun. Be careful not to absorb the magnetic beads.

9. Open the lid of the centrifuge tube, stand and dry for water at room temperature for 5min. Just observe the complete liquid volatilization in the wall and bottom and the smooth surface of the magnetic beads. Pay attention to the drying time should not be too long, which will make the magnetic beads not easy to be eluted.

 10_{N} Add 50-100µL, vortex and mix, incubate 56°C for 5min, the centrifuge tube is placed in the magnetic frame. After the magnetic beads are fully adsorbed in the magnetic frame, the solution is sucked into the new centrifuge tube along the tube wall with a pipette gun, pay attention not to absorb the magnetic beads. The resulting solution is the purified genomic DNA sample and stored in-20°C.

Note:

- 1. The magnetic beads before use with a scroll oscillator.
- 2. The magnetic beads were stored in a 4°C refrigerator.
- 3. Do not freeze the magnetic beads and leave them in a dry state.
- 4. Samples should avoid repeated freezing and thawing, otherwise causing a decrease in extraction volume.

Experimental Result:

Genomic extraction results from 3 mL E. coli bacterial solution.

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ng/ µl	A260/ A280	A260/ A230
160.1	1.92	1.87
184.5	1.94	2.12
157.8	1.88	2.49
188.1	1.86	2.33
165.3	1.83	1.79
165.6	1.89	1.99



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