

MagicPure® Stool and Soil Genomic DNA Kit

Cat. No. EC801

Storage: At room temperature (15-25°C) for one year.

Description

MagicPure® Stool and Soil Genomic DNA Kit provides a simple and fast method to isolate and purify genomic DNA from stool or soil samples. To achieve effective removal of inhibitors and contaminants in solid and liquid samples, unique lysis buffer is designed and magnetic beads are used for specific binding to DNA. The isolated DNA is suitable for various molecular biology general experiments, such as PCR, qPCR and Next Generation Sequencing, etc. This kit is compatible with high-throughput automated nucleic acid purification instruments using magnetic rods.

Features

- · Simple and fast
- · High yield and purity

Starting material

Fresh or frozen stool or soil, avoiding repeated freezing and thawing.

Kit Contents

Component	EC801-11 (50 rxns)
Lysis Buffer 31 (LB31)	50 ml
Lysis Buffer 35 (LB35)	30 ml
Precipitation Buffer 31 (PB31)	12.5 ml
Precipitation Buffer 35 (PB35)	12.5 ml
Binding Buffer 31 (BB31)	10 ml
Clean Buffer 31 (CB31)	36 ml
Wash Buffer31 (WB31)	15 ml
Elution Buffer (EB)	10 ml
Magnetic Stool and Soil Beads	1 ml
Glass Beads	12.5 g

Procedures

Before starting, add indicated volume of 100% isopropanol into BB31 while 100% ethanol into CB31 and WB31 (see the table below). Before use, please make sure BB31 is mixed well.

All magnetic separation steps should be carried out at room temperature Please prepare a 70°C water bath or other heating equipment before starting and prepare 2 ml centrifuge tube.

Component	EC801-11
Binding Buffer 31 (BB31)	30 ml 100% isopropanol
Clean Buffer 31 (CB31)	9 ml 100% ethanol
Wash Buffer 31 (WB31)	60 ml 100% ethanol





Stool genomic DNA extraction

1. Lysis

Weigh 100-300 mg or 100-200 μ l stool sample in a 2 ml centrifuge tube. Add 0.25 g glass beads and 1 ml LB31 [If RNA-free genomic DNA is required, add 20 μ l RNase A (Cat. No. GE101-01) to the sample and incubate at room temperature for 3 minutes]. Vortex the tube sufficiently until the stool sample is thoroughly homogenized and lysed. Then incubate at 70°C for 10 minutes. After pretreatment, centrifuge the tube containing the stool sample at 12,000×g for 5 minutes, and then transfer the supernatant to a new 1.5 ml sterile centrifuge tube.

2. Precipitation

Add 250 μ l PB31 and mix well by vortexing. Then incubate on ice for 5 minutes. Centrifuge at 12,000×g for 2 minutes, and transfer no more than 600 μ l of clear liquid in the middle layer to a 1.5 ml sterile centrifuge tube (After centrifugation, the bottom layer contains precipitates, and the top layer contains a lot of foams. Only the clear liquid in the middle layer is required. If the transferred liquid is still turbid, centrifuge at 12,000×g for 1 minute and transfer the supernatant to a 1.5 ml sterile centrifugal tube).

- 3. Add 500 µl BB31 and mix well. Add 15 µl magnetic beads to the tube and vortex for 5 minutes. Then place the tube on a magnetic stand until the solution becomes clear.
- 4. Discard the supernatant carefully (avoid pipetting the beads). Add 800 μl CB31 and vortex for 5 minutes. Then place the tube on a magnetic stand until the solution becomes clear.
- 5. Discard the supernatant carefully (avoid pipetting the beads). Add 700 μl WB31 and vortex for 3 minutes. Then place the tube on a magnetic stand, until the solution becomes clear.
- 6. Repeat step 5 once.
- 7. Pipette supernatant as thoroughly as possible. Air-dry the uncapped beads at room temperature for 5-10 minutes to volatilize the residual ethanol completely.
- 8. Add 50-100 μl EB. Vortex for 30 seconds and incubate at 65°C for 5 minutes (vortex 3 times during this period). Place the tube on a magnetic stand until the solution becomes clear. Transfer the supernatant carefully into a new sterile centrifuge tube.
- 9. Store the purified gDNA at -20°C.

Soil genomic DNA extraction

1. Lysis

Weigh no more than 250 mg soil sample in a 1.5 ml centrifuge tube. Add 0.25 g glass beads and $600 \mu l$ LB35. Vortex for 15 minutes and then centrifuge at $12,000 \times g$ for 2 minutes. Transfer no more than $400 \mu l$ of supernatant into a 1.5 ml sterile centrifuge tube.

2. Precipitation

Add $200 \,\mu$ l PB35 and mix evenly by vortexing. Then incubate on ice for 5 minutes. Centrifuge at $12,000 \times g$ for 3 minutes, and transfer supernatant to a new 1.5 ml centrifuge tube.

3. The following steps are the same as steps 3-9 in "Stool genomic DNA extraction" section.

Notes

- The stool sample stored in ethanol is recommended to be centrifuged to remove ethanol and washed with sterilized water for 2-3 times before use.
- Beads must be mixed well by vortexing before use.
- Use sterile tubes and pipette tips to avoid contamination from DNase.
- Before extracting, vortex to completely homogenize the sample.
- To ensure high yield, samples should be kept under optimal storage conditions.
- To ensure high extraction purity, avoid pipetting the precipitate while transferring supernatant.

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