TIANGEN®

Magnetic Soil And Stool DNA Kit

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Magnetic Soil And Stool DNA Kit

Cat.no. 4992736/4992738

Kit Contents

Contents	4992736 (50 preps)	4992738 (200 preps)
Buffer SA	45 ml	120 ml
Buffer SC	5 ml	25 ml
Buffer SH	10 ml	45 ml
Buffer GFA	10 ml	30 ml
Buffer RD	24 ml	90 ml
Buffer PWD	20 ml	2×40 ml
Buffer TB	15 ml	30 ml
1 mm Grinding Beads	15 g	60 g
MagAttract Suspension G	0.5 ml	2×1 ml
Handbook	1	1

Optional Products

Magnetic Stand (Cat. no. OSE-MF-01); RNase A (10 mg/ml)

Storage Conditions

This kit can be stored for 12 months in a dry environment under room temperature (15-25°C). It can be stored for a longer time under the temperature of 2-8°C. If the solution produces precipitation under the temperature of 2-8°C, please place the solution in the kit at room temperature for a period of time before use. If necessary, it can be placed in a 37°C water bath for 10 min to dissolve the precipitation.

Introduction

The kit adopts a unique buffer system, by which the humic acid in soil samples could be completely removed. Special grinding beads are also applied in this kit to effectively process the lysis of various complex components in soil samples, thus ensuring the integrity of extracting genomic DNA from soil. This kit is also suitable for extracting genomic DNA from feces samples.

The DNA recovered by using the kit has few impurities and good integrity, and can be directly used in molecular biology downstream experiments such as PCR, enzyme digestion, etc.

Features

Wide application: It is suitable for the extraction of various soil environmental samples such as flower bed soil, flowerpot soil, farmland soil, mountain forest soil, silt, red soil, black soil, dust, etc, and is also suitable for the extraction of fecal samples.

Convenient operation: The experimental operation can be completed in short time.

High purity: Combined with magnetic bead purification, the extracted DNA has high purity and can be directly used in downstream experiments.

Notes **Please read the notes before using this kit**

1. Fresh soil sample could have a higher yield. Proper storage conditions of different soil samples should be checked before sampling.
2. Be careful not to touch the precipitate in any supernatant collection steps, or else the product purity would be affected.
3. Appropriate volume of isopropanol need to be added to Buffer GFA, Buffer RD and Buffer PWD as indicated on the bottle tag before use.
4. Excessive amount of gDNA could inhibit the PCR reaction, so please dilute the gDNA to an optimal concentration in such a circumstance.
5. Before use, check whether there is precipitation in Buffer SC. If there is precipitation, please heat it at 37°C until it is completely dissolved before using.
6. There may be RNA residues in stool samples. If RNA needs to be removed, please prepare RNase A solution.

Protocol

Please add isopropanol to Buffer GFA and ethanol (96-100%) to Buffer RD and Buffer PWD as indicated on the bottle tag.

1. Sample treatment

1) Soil sample treatment:

Add 0.25-0.5 g of sample into a 2 ml centrifuge tube, and add 500 µl of Buffer SA, 100 µl of Buffer SC and 0.25 g of grinding beads. Vortex for 15 min until the sample is evenly mixed or evenly mixed using TGrinder H24 Tissue Homogenizer (OSE-TH-01) (6 M/S for 30 sec at intervals of 30 sec for 2 cycles). Centrifuge at 12,000 rpm (~13,400 × g) for 1 min and transfer the supernatant (about 500 µl) to a new 2 ml centrifuge tube.

Notes: For some samples with low yield or for the extraction of fungal genomes, it is suggested to mix evenly with vortex or tissue homogenizer, and then heat the sample at 70°C for 15 min to improve the lysis efficiency.

2) Stool sample treatment:

Add 0.25-0.5 g of sample to 2 ml centrifuge tube, or transfer 200 µl to the centrifuge tube if it is liquid sample. Add 500 µl Buffer SA, 100 µl Buffer SC and 0.25 g grinding beads (RNA may be residual in stool sample. To remove RNA, it is recommended to add 10 µl RNase A (self-provided)) and mix evenly with vortex or tissue homogenizer, then heat at 70°C for 15 min to improve lysis efficiency. Centrifuge at 12,000 rpm (~13,400×g) for 1 min and transfer the supernatant (about 500 µl) to a new 2 ml centrifuge tube.

Notes: For gram-positive bacteria that are difficult to break the cell wall, the temperature can be increased to 95°C to promote lysis.

2. Add 200 µl Buffer SH, mix well, and vortex for 5 sec, and stand at 4°C for 10 min.
3. Centrifuge at 12,000 rpm (~13,400×g) for 3 min, and transfer the supernatant to a new 2 ml centrifuge tube, then add 500 µl Buffer GFA **(ensure that isopropanol has been added before use)**, and mix evenly upside down.

Notes: Do not touch precipitate when transferring supernatant, otherwise DNA purity may be reduced.

4. Add 10 µl of MagAttract Suspension G and shake to mix well for 5 min.

   Notes: To ensure that the magnetic beads are completely re-suspended, shake to mix before use.

5. Place the centrifuge tube on the magnetic stand for 30 sec. After the magnetic beads are completely attached, carefully remove the liquid.
6. Remove the centrifuge tube from the magnetic stand, and add 700 µl of Buffer RD **(ensure that ethanol(96-100%) has been added before use)**, then shake to mix well for 5 min.
7. Place the centrifuge tube on the magnetic stand for 30 sec. After the magnetic beads are completely attached, carefully remove the liquid.
8. Remove the centrifuge tube from the magnetic stand, and add 700 µl of Buffer PWD **(ensure that ethanol(96-100%) has been added before use)**, then shake to mix well for 3 min.
9. Place the centrifuge tube on the magnetic stand for 30 sec. After the magnetic beads are completely attached, carefully remove the liquid.
10. Repeat Step 8 and 9 once.
11. Place the centrifuge tube on a magnetic stand and dry at room temperature for 5-10 minutes.

    Notes: Ethanol residue will inhibit the subsequent enzyme reaction, so make sure that the ethanol volatilizes completely when drying. But don't over-dry the beads as it will be difficult to elute DNA.

12. Remove the centrifuge tube from the magnetic stand, and add 50-100 µl of Buffer TB. Shake to mix, and incubate at 56°C for 5 min, and mix it upside down for 3 times, 3-5 times each time.
13. Place the centrifuge tube on the magnetic stand for 2 min, after the magnetic beads are completely attached, carefully transfer the DNA solution to a new centrifuge tube, and store it in proper conditions.