TIANamp Soil DNA Kit

For isolation of genomic DNA from soil

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TIANamp Soil DNA Kit
(Spin Column)

Cat. no. 4992288

Kit Contents

Contents	4992288 50 preps
Buffer SA	45 ml
Buffer SC	5 ml
Buffer HA	15 ml
Buffer HB	15 ml
Buffer GF	70 ml
Buffer PWS	15 ml
Buffer TE	15 ml
1 mm Grinding Beads	15 g
Spin Columns CB3	50
Collection Tubes 2 ml	50
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Storage

TIANamp Soil DNA Kit should be kept in dry place and can be stored at room temperature (15-25°C) for up to 12 months. For longer storage, the kit can be stored at 2-8°C. If a precipitate has formed in Buffer under 2-8°C, please place the buffer at room temperature or warm at 37°C for 10 min to dissolve the precipitate.

Introduction

TIANamp Soil DNA Kit uses a unique buffer system, by which the humic acid in soil sample could be completely removed. 1 mm Grinding Beads are also applied in this kit to process the lysis of components of soil sample in order to guarantee the integrity of gDNA.

The genomic DNA isolated by this product is highly pure and has high integrity, so it serves as an excellent template for downstream molecular biological experiments like PCR analysis and restriction enzyme digestion.

Product Features

Wide application:

Good for gDNA isolation from flower bed soil, potting soil, farmland soil, forest soil, sludge, red soil, black soil, dust and many other kinds of soil samples.

Convenient operation:

The whole experimental procedure could be finished within a relatively short time.

High purity:

With the application of spin column method, gDNA isolated by this kit have high purity and is good for downstream experiments.

Important Notes (Please read before use)

1. Fresh soil sample could have a higher yield. Best storage conditions of different soil samples should be checked before sampling.
2. All centrifugation steps should be carried out in a conventional table-top microcentrifuge at room temperature (15-25°C).
3. Avoid pipetting precipitate in any supernatant collection steps, or else the column would be blocked and the product purity would be affected.
4. Appropriate volume of ethanol need to be added to Buffer PWS as indicated on the bottle tag before use.
5. Excessive amount of gDNA could inhibit the PCR reaction, so please dilute the gDNA to an optimal concentration in such a circumstance.

Protocol

Ensure that Buffer PWS have been prepared with appropriate volume of ethanol as indicated on the bottle tag and shake thoroughly.

1. Add 750 µl Buffer SA and 0.25 g Grinding Beads to a 2 ml microcentrifuge tube.
2. Add 0.25 g soil sample to the 2 ml microcentrifuge tube, mix by vortex for 15 sec.
3. Add 60 µl Buffer SC to the sample and mix by vortex for 10 min.
   Note: Check if there is any precipitate in Buffer SC before use, if there is, please dissolve the precipitate by warming at 37°C.
4. Centrifuge at 12,000 rpm (~13,400 × g) for 1 min, transfer the supernatant (around 500 µl) to a new 2 ml microcentrifuge tube.
5. Add 250 µl Buffer HA, mix by vortex for 5 sec, and incubate the tube at 4°C for 5 min.
6. Centrifuge at 12,000 rpm (~13,400 × g) for 1 min, transfer the supernatant to a new 2 ml microcentrifuge tube, add 200 µl Buffer HB and mix, incubate the tube at 4°C for 5 min.
   Note: Avoid pipetting precipitate when transferring supernatant, or else the purity of product would be affected.
7. Centrifuge at 12,000 rpm (~13,400 × g) for 1 min, transfer the supernatant to a new 2 ml microcentrifuge tube, add 1,200 µl Buffer GF and mix by inverting.
   Note: Avoid pipetting precipitate when transferring supernatant, or else the purity of product would be affected.
8. Transfer 700 µl solution from step 7 to a Spin Column CB3 (place the column in a collection tube), centrifuge at 12,000 rpm (~13,400 × g) for 30 sec, discard the flow-through, put the column back to the collection tube.
   Note: The capacity of Spin Column CB3 is 700 µl, if the sample volume exceeds 700 ul, centrifuge successive aliquots in the same column. Discard the flow-through after each centrifugation.
9. Add 500 µl Buffer PWS **(Ensure that ethanol (96-100%) has been added)** to the Spin Column CB3, centrifuge at 12,000 rpm (~13,400 × g) for 30 sec, discard the flow-through, put the column back to the collection tube.
10. Add 500 µl 70% ethanol to the Spin Column CB3, centrifuge at 12,000 rpm (~13,400 × g) for 30 sec, discard the flow-through, put the columnback to the collection tube.
11. Centrifuge at 12,000 rpm (~13,400 × g) for 2 min, discard the flowthrough.
12. Incubate the Spin Column CB3 at room temperature (15-25°C) for several minutes to completely dry the residual washing buffer in the column. Place the Spin Column CB3 in a new clean microcentrifuge tube, and pipet 50-100 µl Buffer TE directly to the center of the membrane. Incubate at room

temperature (15-25°C) for 2-5 min, and then centrifuge for 2 min at 12,000 rpm (~13,400 × g), collect the flowthrough to the microcentrifuge tube.

Note: In order to get a high yield, flow-through could be pipetted back to the Spin Column CB3 and incubated at room temperature (15-25°C) for 2 min, then centrifuge again at 12,000 rpm (~13,400× g) for 2 min. The pH value of elution buffer has a great impact on elution efficiency, we suggest that the pH value should be within the range of 7.0-8.5 if distilled water is used as elution buffer. Low pH value (pH<7) would significantly reduce the efficiency of elution. DNA product should be stored at -20°C to avoid the degradation.

Analysis of DNA concentration and purity

Size of gDNA extracted by this kit is related with the sample storage condition, shearing force during operation and some other factors. Purified DNA can be analysis by electrophoresis gel and UV-Spectrophotometer.

DNA has a significant peak at OD_{260}. An OD_{260} of 1 corresponds to a 50 μg/ml of dsDNA solution or a 40 μg/ml of ssDNA solution.

$OD_{260/280}$ ratio value should be within 1.7-1.9. If ddH_2O is used to elute DNA, the ratio value would be lower, since the pH value and the existence of ion could affect the absorption value.