

High-Q™-Spin-Column Soil Microbiome DNA Purification Kit

Ordering info

TBK0417, 50 reactions

Description

The High-Q™ Spin-Column Soil Microbiome Genomic DNA Purification Kit is an easy-to-use, silica-membrane-based system for DNA purification from different soil types. This kit allows the isolation of high molecular weight genomic DNA from various microorganisms, including Gram-positive and Gram-negative bacteria, archaea, and fungi.

The High-Q™ Spin-Column Soil Microbiome Genomic DNA Purification Kit includes an optimized lysis buffer and a precipitation buffer specifically designed for difficult soil types that contain a high concentration of humic acid. The use of High-Q™ Soil DNA Spin Columns and the PCR inhibitors removal buffer ensures high-quality DNA, suitable for downstream application.

Features

- High yield and purity, 0.5-15 µg depending on sample material, A260/A280 1.8 -2.0; A260/A230 1.4 -1.8.
- Eliminates PCR inhibitors including humic acid.
- Easy and cost-effective protocol.

Applications

DNA obtained is suitable for downstream molecular biology applications such as PCR techniques, microbiome analysis (NGS) and hybridization methods.

Kit Components

Components	TBK0417
High-Q™ Soil DNA Spin Column	50
Collection Tube	100
Bead Tubes	50
Proteinase K ^a	30 mg
Proteinase K Resuspension Buffer	2 mL
Microbiome Soil-1 Buffer	50 mL
Microbiome Soil-2 Buffer	12 mL
Microbiome Soil-3 Buffer	45 mL
Microbiome Soil-4 Buffer	27 mL
WB2 Buffer	45 mL
Elution Buffer	6 mL

Order Info Kit Components: High-Q™ Soil DNA Spin Column (TBM0011) | Collection Tubes (TBM0020) | Bead Tubes (TBR0128) | Microbiome Soil-1 Buffer (TBK0417-1) | Microbiome Soil-2 Buffer (TBK0417-2) | Microbiome Soil-3 Buffer (TBK0417-3) | Microbiome Soil-4 Buffer (TBK0417-4) | WB2 Buffer (TBB0512) | Elution Buffer (TBB0510). Proteinase K (TBZ0305) | Proteinase K Resuspension Buffer (TBB0546).

Before its use:

^a Add 1.5 mL Proteinase K Resuspension Buffer and mix well.

Storage

Store the kit at 25°C. Store Proteinase K at -20°C.

Quality Control

Soil DNA isolation is checked by: integrity (agarose gel electrophoresis), quantity and quality A260/A280 1.8 - 2.0; A260/A230 1.4 -1.8.

PROTOCOL

1. Place **200-300 mg of soil sample** in a Beads Tube provided. Add **850 µL Microbiome Soil-1 Buffer** and mix by vortex.

If the soil sample is very wet, transfer it to a clean tube and centrifuge for 1 minute at 10,000g to remove excess liquid. Very dry material can absorb large volumes of lysis buffer. In this case, either reduce the amount of sample material or add additional lysis buffer.

2. Add **25 µL of Proteinase K** (20 mg/mL). Incubate at 70 °C for 10 minutes.

3. Homogenize by continuously shaking horizontally at maximum speed in a vortex for 15 minutes.

Ensure that the vortex adapter enables horizontal agitation. The agitation with vertically oriented tube adapters does not produce a good yield. A mixer mill, such as GenoGrinder 2010, Fastprep-24®, or similar could be used. The lysis time could change by increasing it to 10, 20, or 30 minutes, but less shaking time avoids shearing of DNA.

4. Centrifuge at 14,000 g for 2 minutes and transfer carefully the supernatant to a clean 1.5 mL tube.

Expect between 500-600 µL of supernatant.

5. Add **200 µL Microbiome Soil-2 Buffer**. Mix by vortex.

6. Centrifuge at 14,000 g, 2 minutes and carefully transfer 600 µL of supernatant to a clean 1.5 mL microcentrifuge tubes avoiding touching the pellet.

7. Add **900 µL Microbiome Soil-3 Buffer** and vortex briefly.

8. Transfer up **750 µL mixture** to a High-Q™ Spin Column placed into a Collection Tube.

9. Centrifuge at 10,000 g for 1 minute. Remove the flow-through and place back the High-Q™ Spin Column into the Collection Tube. Repeat steps 8 and 9 until all the sample has been processed.

Ensure that the entire mixture has passed into the collection tube. If sample remains in the column, centrifuge again at 10,000 g for 1 minute.

10. Add **500 µL Microbiome Soil-4 Buffer**.

11. Centrifuge at 10,000 g for 1 minute. Remove the collection tube and place back the High-Q™ Spin Column into a clean Collection Tube.

12. Add **700 µL WB2 Buffer**.

13. Centrifuge at 10,000 g for 1 minute. Remove the flow-through and place back the High-Q™ Spin Column into the Collection Tube.

14. To dry High-Q™ Spin Column and eliminate residual ethanol, centrifuge again at 10,000 g for 2 minutes.

15. Place the High-Q™ Spin Column into a clean 1.5 mL Tube. Add **50-100 µL prewarmed Elution Buffer**.

Prewarm Elution Buffer at 70°C.

16. Incubate at room temperature, 2 minutes. Centrifuge at 10,000 g for 1 minute to elute purified DNA.

17. Check DNA quantity by spectrophotometry and quality on agarose electrophoresis gel. Store DNA at -20°C.