FastBeat Soil DNA Kit (Bead Beating)

Product Number: DNK4501

Shipping and Storage

All reagents, when store in indicated temperature, are stable for 9 months.

Components

Component	Storage	DNK4501 50 Preps
Bead Tube	RT	50
Buffer SP	RT	50 ml
Buffer MT	RT	6 ml
Buffer PPS	RT	13 ml
Buffer IRS	RT	15 ml
Buffer PQ	RT	35 ml
Buffer WB	RT	13 ml
Buffer EB	RT	10 ml
DNA Bind Columns	RT	50

Description

The FastBeat Soil DNA Kit quickly and efficiently isolates PCR-ready genomic DNAdirectly from soil samples in less than 40 minutes. Designed for use with Beads-Beating device such as the FastPrep® Instruments from MP Biomedicals, soil organisms population are easily lysed within 40 seconds. Samples are placed into 2.0 ml tubes containing 3 kinds of beads, a mixture of ceramic and silica particles designed to efficiently lyse all soil organisms including historically difficult sources such as eubacterial spores and endospores, gram positive bacteria, yeast, algae, nematodes and fungi.

The kit uses a novel and proprietary method to remove high humic acid content including difficult soil types such as compost, sediment, and manure.The isolated DNA has a high level of purity allowing for more successful PCR amplification of organisms from the sample. PCR analysis has been performed to detect a variety of organisms including bacteria (e.g. Bacillus subtilis, Bacillus anthracis), fungi (e.g. yeasts, molds), algae and Actinomycetes (e.g. Streptomyces).

Note

The fill volume in the bead tube after the addition of the Sodium Phosphate and Buffer MTs to the sample should allow sufficient air space in the sample tube for efficient FastPrep® Instrument processing. We recommends using 500 mg of starting material as long as there is between 250 - 500 µl of empty space in the tube. Sample loss or tube failure may result from overfilling the bead tube. The bead tube caps must be secure, but not over-tightened, to prevent sample leakage. If the sample is too large for processing in a single tube, divide the sample and process using multiple tubes. The kits have been rigorously tested in the FastPrep® Instrument. A single 40 second run at a speed setting of 6.0 in the FastPrep® Instrument is sufficient to lyse almost all samples. If the user experimentally determines that additional processing time is required, the sample should be incubated on ice in the bead tube for at least 2 minutes between successive FastPrep® Instrument homogenizations to prevent overheating the sample and tube.

If you use other bead beater device, please follow instruction manual from manufaturer to set appropriate parameter for good performance.

Protocol(Please read the precautions before the experiment)

Tip:Before the first use, add the indicated amount of ethanol into Buffer PQ bottle, Buffer WB bottle, mix well, and mark the bottle with a check.

Email: sales@mebep.com Website: www.mebep.com
Tel: +86-755-86134126 WhatsApp/Facebook/Twitter: +86-189-22896756

1. Add up to 500 mg of soil sample to a Bead Tube.
2. Add 980µl Buffer SP to sample in Bead Tube. Gentle votex to mix. Add 120 µl Buffer MT.
   Note: Check Buffer MT. If Buffer MT is precipitated, heat solution to 60°C until dissolved before use.
3. Homogenize in the FastPrep® Instrument for 40 seconds at a speed setting of 6.0.
4. Centrifuge at 12,000 x g for 5minutes to pellet debris.
5. Transfer supernatant to a clean 2.0 ml centrifuge tube. Add 250µl Buffer PPS and mix by shaking the tube by hand 10 times. Incubate at 4°C for 5 minutes.
6. Centrifuge tubes at 10,000 x g for 3 minute at room temperature. Avoiding pellet, transfer up to, but no more than, 900 µl of supernatant to a clean 2 ml centrifuge tube.
7. Add 300 µl of Buffer IRS(1/3 volume) and vortex briefly. Incubate at 4°C for 5 minutes.
8. Centrifuge tubes at 10,000 x g for 1 minute at room temperature. Avoiding pellet, transfer the supernatant into a clean 5 ml centrifuge tube.
9. Add 1.5 volumes of Buffer PQ to the cleared supernatant and mix by pipetting.
   Example: To 1100 µl lysate add 1650 µl Buffer PQ. Reduce the amount of Buffer PQ accordingly if less supernatant is recovered. A precipitate may form after the addition of ethanol but this will not affect the procedure.
   Note: 1)Ensure ethanol has been added to Buffer PQ.
   2)It is important to pipet Buffer PQ directly onto the cleared supernatant and to mix immediately.
10. Load approximately 700 µl mixture onto Spin Filter(sitting in collection tube) and centrifuge at 10,000 x g for 1 minute at room temperature. Discard flow through. Load another 700 µl and repeat untill all remaining mixture is loaded on Spin Filter.
    Note: A total of 4-5 loads for each sample processed may be required.
11. Add 600 µl of Buffer WB to Spin Filter and centrifuge at 10,000 x g for 30 seconds at room temperature. Discard flow through. Repeat Step 11 with another 600 µl Buffer WB.
    Note: Ensure ethanol is added to Buffer WB.
12. Centrifuge Spin Filter at 13,000 x g for 2 minute at room temperature to dry the Spin Filter..
13. Carefully place Spin Filter in clean 1.5 ml centrifuge tube. Avoid splashing any Buffer WB onto Spin Filter. Add 100 µl of Buffer EB(Optional: pre-warm the water to 70–90C will increase the DNA yield) to the center of the column membrane. Incubate at room temperature for 3-5 min, and centrifuge at 13,000 x g for 1 min to elute the DNA.
    Note: Use smaller volume(minimum 30µl) of Buffer EBwill obtain higher concentration.
    Optional: Put eluate back to the Spin Column to repeat elution once. This increases concentration of DNA about 10-15%.