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General Information

Qbiogene is a pioneer in developing kits for molecular biology research. We introduced the GENE CLEAN[®] Kits in 1986 and have since been manufacturing products to bring convenience into your research. Our goal is to make your life easier by simplifying the complexities of lab work.

Technical Support and Ordering Information

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FastDNA[®] SPIN Kit for Soil

Catalog # 6560-200

50 preps

- Purifies PCR-ready Genomic DNA from Soil Samples
- For use with the FastPrep[®] Instrument

Shipping and Storage:

The FastDNA[®] SPIN Kit for Soil is shipped and stored at ambient temperature.

Revision # 6560-999-1D06

FastDNA[®] SPIN Kit for Soil Protocol

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Call (800) 424-6101 for Technical Support

**Kit Components
6560-200 (50 preps)**

Name	Volume	Cat #
Lysing Matrix E	100	6914-050
PPS (Protein Precipitation Solution)	25ml	6540-403
Binding Matrix (DNA Binding Matrix Solution)*	66ml	6540-408
SEWS-M (Salt/Ethanol Wash Solution)**	12ml	6540-405
DES (DNA Elution Solution-Ultra Pure Water)	20ml	6540-406
Sodium Phosphate Buffer	60ml	6560-205
BBS gel loading dye	200ul	6540-407
MT Buffer	8ml	6511-202
SPIN Filters	50	6560-210
Catch Tubes	50	6560-211

All references to user supplied reagents are italicized and bolded in the protocol for your convenience.

*Contains Guanidine Thiocyanate. Use with proper caution.

**Protocol for preparation of SEWS-M (salt/ethanol wash solution, DNase-free): Add 100 ml ethanol to the bottle labeled SEWS-M, which already contains 12 ml of DNase-free salt solution, to make a total of 112 ml. Shake and store tightly capped bottle at room temperature.

‡In case a precipitate is seen, heat bottle in 45-55°C water bath and allow to cool to room temperature.

Introduction

The **FastDNA® SPIN Kit for Soil** is designed to extract PCR-ready Genomic DNA from soil samples in less than 30 minutes. The rapid DNA extraction method precludes the use of harmful organic solvents such as phenol and chloroform. The kit enables the extraction of genomic DNA from all bacteria, fungi, plants and animals in a soil community.

The kit consists of three general components:

1. Lysing Matrix

The **Lysing Matrix E tubes** contain a mixture of ceramic and silica particles designed to efficiently lyse all microorganisms including historically difficult sources such as eubacterial spores and endospores, gram-positive bacteria, yeast, algae, nematodes, and fungi.

2. Homogenization Reagents

The **MT Buffer** and **Sodium Phosphate Buffer** have been carefully selected and prepared to enable complete sample homogenization and protein solubilization. The reagents enable extraction of genomic DNA with minimal RNA contamination.

3. DNA Purification and Elution Reagents

A **GENECLEAN®** procedure is then used to purify the genomic DNA. The procedure purifies DNA with a proprietary silica matrix and eliminates contaminants that inhibit subsequent reactions.

- * **Binding Matrix** Suspension
- * **SEWS-M** (Salt Ethanol Wash)
- * **DES** (DNA Elution Solution Ultra-pure water)

Notes

How the System Works

The **FastPrep[®] Instrument*** shakes a tube up and down at very high speeds. The rotor holds 12 x 2 ml tubes enabling 12 samples to be processed simultaneously.

During the processing the 2ml tubes that contain the Lysing Matrix sample also contain a chaotropic DNA stabilizing solution which is a proprietary mixture of detergents and salts. The detergents have been found to serve two functions. One, they contribute to inactivate nucleases. Two, they provide lubrication during the lysing step to control the degree of shearing of the DNA.

Summary of the FastPrep[®] System

The **FastPrep[®] System**, which includes both the **FastPrep[®] Instrument** and **FastDNA[®]** and **FastRNA[®]** kits, has the ability to lyse cells with minimal shearing of the nucleic acids. The procedure eliminates the major concerns in isolation of nucleic acids from cells that are difficult to lyse without enzymes, manual grinding, or homogenizing. It is these laborious and time consuming lysing steps which allow nucleases to act and can make nucleic acid isolation a chore. The **FastPrep[®] System**, by use of highly energetic mechanical means and careful choice of reagents, disrupts whole tissues, lyses cells, and stabilized nucleic acid from any source, thus eliminating the need for lysing enzymes or grinding and homogenizing equipment.

* *Patents pending*

Protocol

Sample Processing:

1. Add up to 500mg of soil to **Lysing Matrix E Tube**.

Due to the vigorous motion of the **FastPrep[®] Instrument**, a significant pressure buildup is observed in the tube. The same and the **Lysing Matrix** should not exceed more than 7/8 of the tube in volume. Leaving space in the tube also improves chances for better homogenization. [See Note 1, page 7.]

Lysis:

Add **978µl Sodium Phosphate Buffer** and **122µl MT Buffer**. [See Note 1, page 7.]

2. Secure tubes in **FastPrep[®] Instrument** and process for 30 seconds at speed 5.5.
3. Centrifuge **Lysing Matrix E Tubes** at 14,000 xg for 30 seconds.[See Note 1, page 7.]
4. Transfer supernatant to a clean tube. Add **250µl PPS** reagent and mix by shaking the tube by hand 10 times.
5. Centrifuge at 14,000 xg for 5 minutes to pellet precipitate. Transfer supernatant to a clean 15ml tube. (Resuspend **Binding Matrix Suspension** before use.) Add 1ml **Binding Matrix Suspension** to the supernatant.
6. Place on a rotator or invert by hand for 2 minutes to allow binding of DNA to matrix. Place tube in a rack for 3 minutes to allow settling of silica matrix.
7. Remove 500µl of supernatant being careful to avoid settled **Binding Matrix**. Discard supernatant. Resuspend **Binding Matrix** in the remaining amount of supernatant. Transfer approximately 600µl of the mixture to a **SPIN[™] Filter** and centrifuge at 14,000 xg for 1 minute. Empty the catch tube and add the remaining supernatant to **SPIN[™] Filter** and spin again.
8. Add **500µl SEWS-M** to the **SPIN[™] Filter** and centrifuge at 14,000 xg for 1 minute. Decant flow-through and replace **SPIN[™] Filter** in Catch tube. Centrifuge at 14,000 xg for 2 minutes to “dry” the matrix of residual **SEWS-M** wash solution

9. Remove **SPIN™ Filter** and place in fresh kit-supplied **Catch Tube**. Air dry the **SPIN™ Filter** for 5 minutes at room temperature.

10. Add **50µl DES** (DNase/Pyrogen Free Water) and gently stir matrix on filter membrane with a pipette tip or vortex/finger flip to resuspend the silica for efficient elution of the DNA. Centrifuge at 14,000 xg for 1 minute to transfer eluted DNA to **Catch Tube**. DNA is now application-ready.

Notes

Please Read Before Conducting Any FastPrep® DNA Extractions

1. Note 1

Important: The volumes are calculated to leave an air space of approximately 0.25cc. If less air space is present there is a likelihood of sample loss due to tube failure or deformation around the cap allowing sample to bubble out. Sample loss is caused by an increase in pressure due to temperature rise during **FastPrep®** runs. The presence of 0.25cc of air space in the tube is sufficient to prevent sample loss during routine **FastPrep®** runs.

2. Note 2

Extending spin to 15 minutes can enhance elimination of excessive debris from large samples, or from cells with complex cell walls.

Note: Please check that tubes are balanced by weight and that the bottom or side of the tubes will not scrape the wall of your microcentrifuge as this will cause rapid loss of sample.