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BIO BASIC INC.

96-Well Plate Plant Genomic DNA Mini-Preps Kit

BS8361

Version 5.0
ISO9001 Certified

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96-Well Plate Plant Genomic DNA Mini-Preps Kit

Product information for BS8361:

Kit Contents

Component	BS8361, 2 plates
Universal Buffer PCB	130 ml
Universal Buffer BD	50 ml
Universal PW Solution	72 ml
Universal Wash Solution	30 ml
TE Buffer (pH 8.0)	40 ml
EZ-10 96 Well Binding Plate	2
Deep Well Collection Plate	6
96 Well Storage Plate	2
Sealing film	8
Protocol	1

Note 1: Universal Buffer BD contains chaotropic salt; avoid contact with skin and eyes.
Note 2: Universal PW Solution and Universal Wash Solution are supplied as concentrates. Add **48 ml isopropanol** to **72 ml Universal** PW Solution and **90 ml ethanol** (96-100%) for **30 ml Universal Wash Solution** before use to obtain a working solution.

Storage and Stability

96 Well Plates and all buffers should be stored dry, at room temperature (15-25°C) and kit is stable for 1 year under these conditions.

Introduction

The kit provides a rapid and convenient high throughput technique for mini preparation of high quality genomic DNA from various plants species. DNA of plant lysate is selectively absorbed on each column of the plate and other impurities such as proteins, salts are eliminated from the column. Each column can absorb up to 20 µg of DNA. No phenol extraction and no ethanol precipitations are required. Purified plant genomic DNA is 20-40 kb, and is suitable for downstream applications such as PCR, Real-time RT PCR, Southern blotting, Microsatellite analysis, AFLP, RFLP, and RAPD.

Features:

- ✓ **Fast and Simple.** Using a rapid spin and high throughput format, the entire procedure takes less than 2 hours.
- ✓ **High purity of DNA.** OD_{260}/OD_{280} of purified DNA is generally >1.8.
- ✓ **Compatible** with many downstream applications such as PCR, restriction digestions, real-time PCR, multiplex PCR, RAPD, RFLP, AFLP, Southern Blotting and microsatellite analysis.

- ✓ **Suitable** for a wide variety of plant species and tissue types including some very recalcitrant specimens.

Materials Supplied by User:

Microcentrifuge capable of at least 6,000 × *g*
Pipets and pipet tips
Vortexer
Chloroform
Isopropanol
Ethanol (96 – 100%)
RNase A (20 mg/ml, Optional for RNA-free DNA)
Water bath for heating at 65°C
β-mercaptoethanol

Procedures

1. Grind 100 mg fresh plant tissue (or 20 mg dry plant tissue) to fine powder in liquid nitrogen; transfer the powder to a **Deep Well Collection Plate**.

2. Add 600 µl Buffer PCB and 12 µl of β-mercaptoethanol to the sample. Seal the **Deep Well Collection Plate** with sealing film, and mix thoroughly by vortexing. Incubate at 65°C for 25 min.

   Note: If RNA-free genomic DNA is required, add 20 µl RNase A (20 mg/ml), mix by vortexing, and incubate for 2 min at room temperature before continuing with step 3.

3. Add 0.6 ml of chloroform to each well. Seal the

Deep Well Collection Plate with sealing film, and mix well by vortexing. Centrifuge at 5,700 x g for 2 minutes. Carefully transfer the supernatant (400 µl) to a new clean **Deep Well Collection Plate**.

4. Add 200 µl Universal Buffer BD. Seal the **Deep Well Collection Plate** with sealing film, and mix thoroughly by vortexing.

   Note: If a gelatinous material appears at this step, incubate at 70°C for 10 min.

5. Add 200µl ethanol (96-100%). Seal the **Deep Well Collection Plate** with sealing film, and mix thoroughly by vortexing.

   Note: If a gelatinous material appears at this step, vigorously shaking or vortexing is recommended.

6. Transfer the mixture from step 5 (including any precipitate) into **EZ-10 96 Well Binding Plate** placed in a new **Deep Well Collection Plate**. Centrifuge at 5,000 *x g* (6,000 *rpm*) for 1 min. Discard the flow-through.

7. Add 500 µl Universal PW Solution, and centrifuge for 1 min at 5,000 *x g* (6,000 *rpm*). Discard the flow-through.

   Note: Check the label to ensure PW Solution was diluted with isopropanol.

8. Add 500 µl Universal Wash Solution, and centrifuge for 1 min at 5,000 *x g* (6,000 *rpm*). Discard the flow-through.

Note: Check the label to ensure Wash Solution was diluted with ethanol.

9. Place the empty **EZ-10 96 Well Binding Plate** in the **Deep Well Collection Plate** and centrifuge for an additional 2 min at 5,000 *x g* (6,000 *rpm*) to dry the EZ-10 96 Well Binding Plate membrane. Discard flow-through.

   Note: It is important to dry the membrane of the **EZ-10 96 Well Binding Plate**, since residual ethanol may interfere with subsequent reactions. This centrifugation step ensures that no residual ethanol will be carried over during the following elution.

10. To elute, place a **96 Well Storage Plate** on top of a **Deep Well Collection Plate**, and then place the **EZ-10 96 Well Binding Plate** on the top of a **96 Well Storage Plate.** Add 50 µl TE Buffer directly onto the center part of the **EZ-10 96 Well Binding Plate** membrane. Incubate at room temperature for 1 min, and then centrifuge for 1 min at 5,000 *x g* (6,000 *rpm*) to elute the DNA.

    Note 1: Warm the TE Buffer to 60°C will increase the elution efficiency.

    Note 2: Elution with more than 50 µl (e.g. 200 µl) increases the DNA yield, but the concentration will be lower.

    Note 3: For maximum DNA yield, repeat elution once as described in this step.

Note 4: For maximum DNA concentration, use the eluate in the microcentrifuge tube for the second elution step.

Other Kits Available

EZ-10 Spin Column Plasmid DNA Mini-Preps Kit
BS413 (50 Preps),
BS414 (100 Preps)
BS614 (250 Preps)

EZ-10 Spin Column PCR Products Purification Kit
BS363 (50 Preps)
BS364 (100 Preps)
BS664 (250 Preps)

EZ-10 Spin Column DNA Gel Extraction Kit
BS353 (50 Preps)
BS354 (100 Preps)
BS654 (250 Preps)

PRODUCTS ARE INTENDED FOR BASIC SCIENTIFIC RESEARCH ONLY!
NOT INTENDED FOR HUMAN OR ANIMAL USE!

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