AbraMag® Genomic DNA Purification Magnetic Bead Kit
Plant Material gDNA Modification

1. Intended Use

The plant material gDNA technical bulletin for the AbraMag® Genomic DNA Purification Magnetic Bead Kit (PN 555020) is intended to detail the steps for modifying the existing kit for use in purifying genomic DNA from various plant materials. Because this modified protocol usesa greater amount of several reagents than the standard protocol, the kit will yield only approximately 40 assays instead of 100.

2. Materials Required

Disposable gloves and other protective equipment
Micropipettes with disposable plastic filter tips
1.5 mL sterile plastic microcentrifuge tubes
4°C refrigerator
-20°C freezer
96-100% Ethanol
Tissue disruption equipment (dissection scissors, razor, mortar and pestle with liquid nitrogen, homogenizer etc.)
Balance
Vortexer
Heating block, thermomixer, or water bath capable of 65°C
Magnetic microcentrifuge tube separator, Solo (Gold Standard Diagnostics PN 472270) or Multi-6 (Gold Standard Diagnostics PN 472260) or similar
Minicentrifuge
AbraMag® Genomic DNA Purification Magnetic Bead Kit (PN 555020)

3. Notes and Precautions

- Upon delivery of the kit, remove the RNase A Solution and Proteinase K Solution vials and store at -20°C. Remove the AbraMag® DNA Purification Magnetic Beads and store at 4°C. Do not freeze the magnetic beads solution. All other kit reagents may be stored at room temperature(20-25°C). Do not use after the printed expiration date.
- Use appropriate protective equipment (including but not limited to gloves, lab coats, and safety glasses) when collecting tissues and bacteria. The DNA Binding Solution and DNA Wash Solution 1 contain guanidine hydrochloride, which can be irritating to eyes and skin. Always wear gloves, lab coats, safety glasses, and/or other protective equipment when using these solutions. Refer to Safety Data Sheet for further information.
- Before the first use of the kit, add 96-100% Ethanol to the DNA Binding Solution concentrate, DNA Wash Solution 1 concentrate, and DNA Wash Solution 2 concentrate as specified below. Mark the bottle to indicate that ethanol has been added. Wear gloves when handling the reagents.
- DNA Binding Solution: Add 12 mL 96-100% Ethanol
- DNA Wash Solution 1: Add 42 mL 96-100% Ethanol
- DNA Wash Solution 2: Add 42 mL 96-100% Ethanol
- Before each use, check for any precipitate formation in the solutions. If observed, shake tore-dissolve any precipitates.

4. Procedure

Sample preparation for plants

Homogenize tissue into a fine powder using mechanical methods such as a bead-beater orhomogenizer, mortar and pestle, or liquid nitrogen freezing and grinding. DNA quality is dependent on the quality of the starting material. Keep frozen until extraction.

Extraction procedure

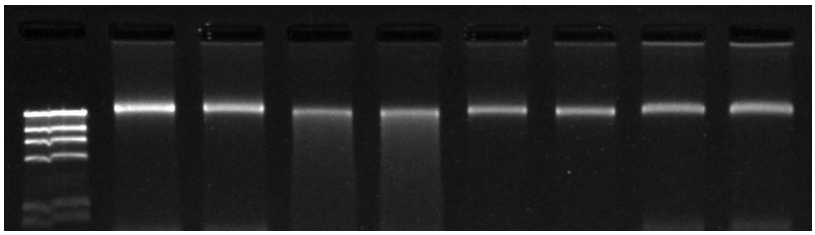
1. Measure up to 10 mg of finely ground plant material into a 1.5 mL microcentrifuge tube. Add500 µL of DNA Lysis Solution and vortex well. Add 20 µL of Proteinase K Solution and vortex again. Ensure that the sample is fully submerged in the solution mix.
2. Incubate the sample at 55°C for 1 hour, vortexing occasionally to suspend the plant material.
3. Centrifuge for 10 minutes at 10,000 x g. Carefully remove the supernatant into a new 1.5 mL microcentrifuge tube, without carrying over any solid material. Discard solid material.
4. *Optional*: Add 20 µL of RNase A Solution to the supernatant and vortex well. Incubate atroom temperature for 10 minutes to degrade plant RNA.
5. In a new 1.5 mL tube, add 400 µL of 96-100% Ethanol. Vortex the AbraMag® DNA Purification Magnetic Beads well to ensure complete resuspension of the beads, and add 20µL of bead solution to the ethanol. Vortex well.
6. Add 300 µL of DNA Binding Solution (prepared with ethanol) to the sample lysate and vortex for 3 seconds. Transfer the sample lysate to the ethanol/bead mix and vortex for 5seconds. Allow the tube to sit at room temperature for ~30 seconds.
7. Place the tube on the magnetic separator for 3 minutes. Leaving the tube on the separator,remove and discard the supernatant using a pipette, without disturbing the beads that havecollected at the magnet.
8. Remove the tube from the magnetic separator and add 600 µL DNA Wash Solution 1 (prepared with ethanol). Vortex briefly to re-suspend the beads and return the tube to the magnetic separator for 2 minutes. Leaving the tube on the separator, remove and discard thesupernatant using a pipette, without disturbing the beads that have collected at the magnet. Repeat wash.
9. Remove the tube from the magnetic separator and add 600 µL DNA Wash Solution 2 (prepared with ethanol). Vortex briefly to re-suspend the beads and return the tube to the magnetic separator for 2 minutes. Leaving the tube on the separator, remove and discard thesupernatant using a pipette, without disturbing the beads that have collected at the magnet. Repeat wash.
10. Pulse spin to remove any wash solution drops remaining on the sides and lid of the tube. Return the tube to the magnetic separator for ~30 seconds. Leaving the tube on the separator, remove and discard any additional supernatant using a pipette, without disturbing the beadsthat have collected at the magnet.
11. Remove the tube from the magnetic separator and add 150 µL DNA Elution Solution. Vortexbriefly to re-suspend the beads.
12. Incubate the sample at 65°C for 10 minutes, vortexing occasionally or using a thermomixer.
13. Pulse spin to remove any condensation from the sides and lid of the tube.
14. Return the tube to the magnetic separator for 2 minutes. Leaving the tube on the separator,transfer the eluate to a new 1.5 mL tube using a pipette. The eluate contains the purified genomic DNA.

5. Evaluation of Results

Results can be evaluated via agarose gel electrophoresis, spectrophotometric or fluorometric readings, PCR evaluation, etc. See the kit User Guide for more details.

6. Performance Data

Plant Type	DNA Yield (µg)
Soybean seed	5.15
Corn seed	1.78
Canola seed	2.13
Linseed	3.00



λHindIII MW ladder | Soy seed | Corn seed | Canola seed | Linseed

7. For ordering or technical assistance contact:

Gold Standard Diagnostics
124 Railroad Drive
Warminster, PA 18974
WEB: www.abraxiskits.com

Phone: (215) 357 3911
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