

Maxwell® 16 LEV Plant DNA Kit

INSTRUCTIONS FOR USE OF PRODUCT AS1420.

Preparation of Plant Leaf Samples for DNA Purification

This Quick Protocol provides instructions for use of the Maxwell® 16 LEV Plant DNA Kit with the Maxwell® 16 Instrument (AS2000) to purify DNA from plant leaf samples. For detailed instructions, including information on instrument setup and troubleshooting, please refer to the *Maxwell® 16 LEV Plant DNA Kit Technical Manual #TM414*, available at:

www.promega.com/protocols/

Preprocessing with a Mechanical Bead-Beating Device

Materials to Be Supplied by the User

- Bead-beating device (e.g., MP Biomedicals FastPrep®-24 Instrument)
 - Sterile, aerosol-resistant pipette tips
 - Microcentrifuge or plate-specific centrifuge
1. Place up to 20mg leaf tissue in the bottom of each tube or well.
 2. Place a bead (or beads, as recommended by manufacturer) into each tube or well.
 3. Add 300µl of Tail Lysis Buffer (TLA) to each tube or well.
 4. Add 10µl of RNase A (optional) to each well.
Note: If processing a large number of samples, prepare sufficient volume of Tail Lysis Buffer and RNase A immediately before use and add 310µl of this cocktail to each sample.
 5. Run the bead-beating device using the time and speed recommended by the manufacturer. Some optimization may be required.
 6. Place the extraction tubes or plates into a centrifuge and spin briefly to remove any solid particulates.
Optional: To reduce foaming of the solution, centrifuge for up to 2 minutes at maximum speed.
 7. Add 300µl of Nuclease Free Water to well #1 of each Maxwell® 16 LEV Plant DNA Kit reagent cartridge. (Well #1 is the well closest to the cartridge label and furthest from the user).
 8. Transfer each plant lysate sample from the extraction tube or plate into well #1 of the Maxwell® 16 Reagent Cartridge. Transfer all liquid and any remaining foam, being careful not to transfer any solid material to the cartridge.

Preprocessing with a Microtube, Pestle, and Liquid Nitrogen

Materials to Be Supplied by the User

- Pellet Pestles (Sigma Aldrich Cat.# Z359947)
 - Liquid nitrogen
 - Sterile, aerosol-resistant pipette tips
 - Microcentrifuge
1. Place up to 20mg leaf tissue in the bottom of a ClickFit MicroTube, 1.5ml.
 2. Add liquid nitrogen to the plant tissue sample. Allow the liquid to evaporate, freezing the sample.
 3. Using a pellet pestle, grind the frozen plant tissue against the tube wall as thoroughly as possible.
 4. Add 300µl of Tail Lysis Buffer (TLA) to each tube.
 5. Add 10µl of RNase A (optional) to each tube.
Note: If processing a large number of samples, combine sufficient volume of Tail Lysis Buffer and RNase A immediately before use and add 310µl of this cocktail to each sample.
 6. Vortex tube briefly (10 seconds).
 7. Place tubes with lysate into a microcentrifuge and spin briefly to remove any solid particulates.
Optional: To reduce foaming of the solution, centrifuge for up to 2 minutes at maximum speed.
 8. Add 300µl of Nuclease Free Water to well #1 of each Maxwell® 16 LEV Plant DNA Kit reagent cartridge. (Well #1 is the well closest to the cartridge label and furthest from the user).
 9. Transfer each plant lysate sample from the extraction tube to well #1 of the reagent cartridge. Transfer all liquid and any remaining foam, being careful not to transfer any solid material to the cartridge.

Maxwell® 16 LEV Plant DNA Kit

INSTRUCTIONS FOR USE OF PRODUCT AS1420.

Cartridge Preparation

1. Place the cartridges to be used in the Maxwell® 16 LEV Cartridge Rack with the label side facing away from the Elution Tube.
2. Press down on the cartridge to snap it into position. Carefully peel back the seal so that all plastic comes off the top of the cartridge. Ensure that all sealing tape and any residual adhesive are removed before placing cartridges in the instrument.

Note: If you are processing fewer than 16 samples, center the cartridges on the cartridge rack.

3. Place an LEV Plunger in well #8 of each cartridge. Well #8 is the well closest to the Elution Tube.

Note: Use only the plungers provided in the Maxwell® 16 LEV DNA Plant Kit.

4. Place 0.5ml Elution Tubes in the front of the Maxwell® 16 LEV Cartridge Rack. Add 50µl of Elution Buffer to the bottom of each Elution Tube.

Notes

- a. If Elution Buffer is on the side of the tube, the elution may be suboptimal.
- b. Use only the 0.5ml Elution Tubes provided in the kit; other tubes may not work with the Maxwell® 16 Instrument.

Figure 1. The LEV plunger is placed in well #8 of the cartridge (the well closest to the Elution Tube), and lysates are placed into well #1 of the cartridge.



Instrument Run on the Maxwell® 16 Instrument (Cat.# AS2000)

1. Refer to the *Maxwell® 16 Instrument (AS2000) Operating Manual #TM295* for detailed information. To run the Plant DNA protocol the Maxwell® 16 firmware version ≥ 4.97 must be installed on the instrument, and the Maxwell® 16 High Strength LEV Magnetic Rod and Plunger Bar Adaptor (Cat.# SP1070) must be used.
2. Follow the instrument run instructions in the *Maxwell® 16 LEV Plant DNA Kit Technical Manual #TM414*.

Ordering and Technical Information

www.promega.com • Phone 608-274-4330
Toll free in USA 800-356-9526 • Fax 608-277-2601