Promega

Product Application

Automated Purification of Genomic DNA from Algae

Purify high quality and high molecular weight genomic DNA from algae using Maxwell® RSC Instrument and Maxwell® RSC PureFood GMO and Authentication Kit

Kit: Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600)

Analyses: UV absorbance, dye-based quantitation, TapeStation and pulsed-field gel electrophoresis

Sample Type(s): *Acutodesmus deserticola* (desert green algae)

Input: 200mg

This protocol was developed by Promega Applications Scientists and is intended for research use only.

Users are responsible for determining suitability of the protocol for their application.

For further information, see Technical Manual TM473 available at: **www.promega.com/protocols**

or contact Technical Services at: **techserv@promega.com**

Materials Required:

- Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600)
- Maxwell® RSC Instrument (Cat.# AS4500)
- Liquid Nitrogen
- Mortar and Pestle
- Microcentrifuge
- LidLocks™ Microcentrifuge Tube Locks, Sorenson BioSciences (Cat.# 11870)

Protocol:

Do NOT vortex the samples. Instead mix by inverting the tubes or pipetting extremely slowly.

Use wide bore pipette tips to minimize DNA shearing. Do NOT use pipette tips that have been cut.

1. Combine all algae into an appropriately sized tube and centrifuge for 5 minutes.
2. Decant all the supernatant from the top of the algae pellet.
3. Pre-cool mortar and pestle with liquid nitrogen.
4. Scrape the algae into the mortar. Add liquid nitrogen and allow to cool about 5 minutes.
5. Grind the sample for 5-10 minutes until the sample is a fine powder.
6. Place the ground sample into a tube and allow the liquid nitrogen to evaporate.
7. Weigh 200mg of sample and place in a 1.5mL microcentrifuge tube.
8. Add 500µl of CTAB buffer and pipet to mix.
9. Secure tube lids (using either screw cap tubes or LidLocks™) and place in a boiling water bath for 5 minutes.
10. Remove the samples and allow them to cool to room temperature.
11. Add 20µl of RNase A Solution and 40µl of Proteinase K to each tube. Mix using very slow pipetting with wide bore pipet tips.

12. Heat the samples at 65°C for 30 minutes.
13. Centrifuge the samples at 16,000 x *g* for 10 minutes at room temperature.
14. Meanwhile, prepare the Maxwell cartridges.
    a. Add 300µl of Lysis Buffer to Well #1 of each Maxwell® cartridge.
    b. Place a plunger into Well #8 of each Maxwell® cartridge.
    c. Add 100µl of Elution Buffer to each elution tube.
15. Transfer 300µl of clear sample lysate to Well #1 of the Maxwell® cartridge.
16. Run the Maxwell® using the program for the Maxwell® RSC PureFood GMO and Authentication Kit.

Results:

DNA was purified from 200mg of *Acutodesmus deserticola* using the above protocol (n=7). Using QuantiFluor® ONE dsDNA System (Cat.# E4871), DNA concentration was measured as 648ng/µl ± SD 98 with a yield of 6.48 µg ± 76 of DNA per 200mg algae pellet.

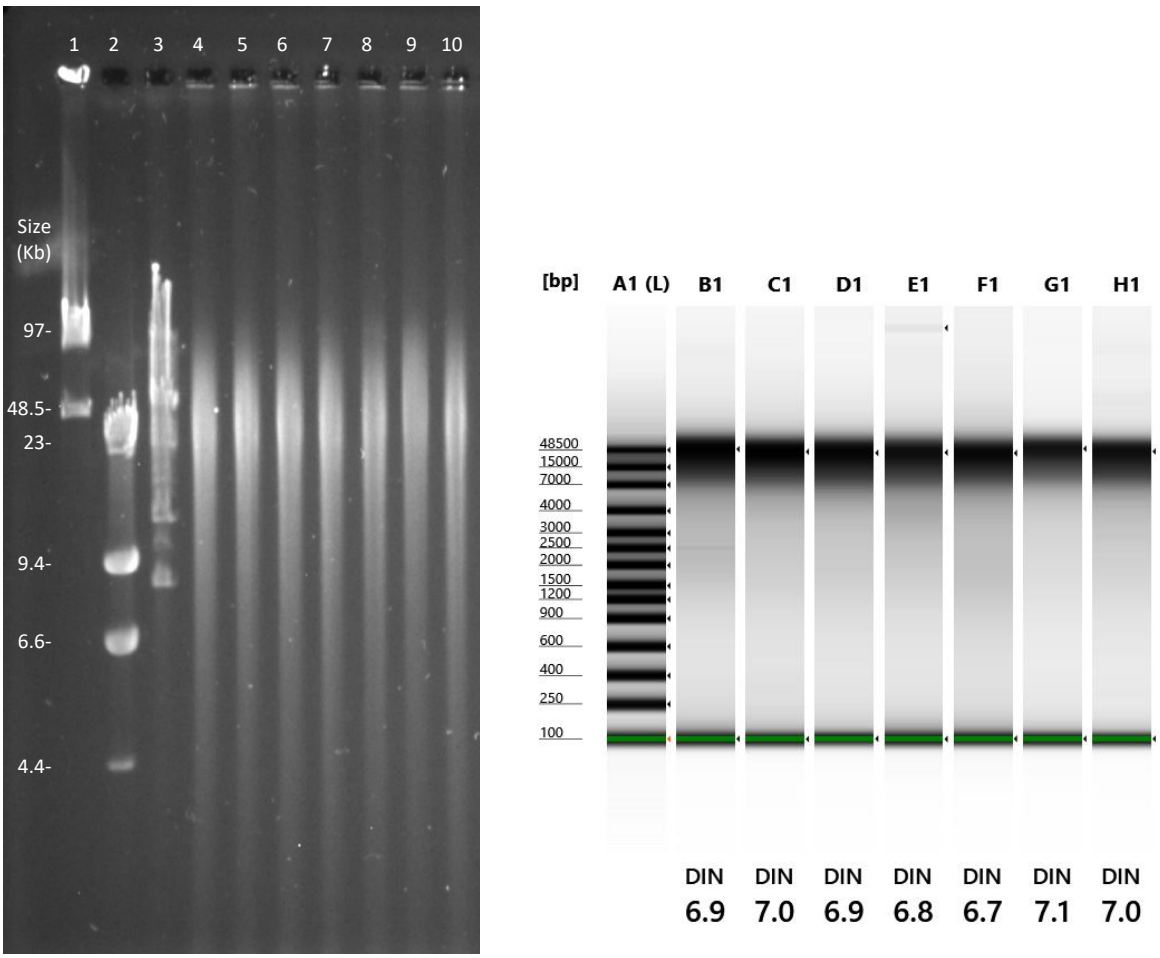

Figure 1. Purified algae DNA size using pulsed field gel electrophoresis (left) and Genomic DNA ScreenTape on an Agilent 4200 TapeStation (right). Samples of high molecular weight DNA were purified using the above protocol with the Maxwell® RSC PureFood GMO and Authentication Kit (Cat.# AS1600) (n=7). **(Left)** 3µl of purified DNA (lanes 4-10), Lambda PFG ladder (New England Biolabs, lane 1), or Lambda DNA/HindIII Marker (Promega, lane 2) were electrophoresed on a 1% agarose gel using a Pippin Pulse Electrophoresis Supply with the protocol for 5-80kb fragments and post-stained with ethidium bromide. **(Right)** 1µl of DNA was analyzed on an Agilent 4200 TapeStation with the Genomic DNA ScreenTape and Reagents. Lane A1, Genomic DNA Ladder; Lanes B1-H1, purified algae gDNA.