The performance evaluation of genomic DNA extraction using the AllEx® Plant DNA/RNA Kit from two bryophyte species (Polytrichum commune and Marchantia polymorpha)

Experimental Conditions

Materials



This image will be updated soon

AllEx®64 Automated Nucleic Acid Extraction System [AEX064] AllEx[®] Plant DNA/RNA Kit [937-048, 937-096]

Sample & Extraction Information

Origin	Bryophyte			
Sample				
	Polytrichum commune (male and female)	Marchantia polymorpha (male and female)		
Target	Genomic DNA			
Sample amount	25 mg			
Elution volume	100 µl			
Extraction protocol	Plant-P3			
Operating time	28' 26''			

Protocol

AllEx[®] Plant DNA/RNA Kit

* For more details and methods, please refer to the manual of AllEx® Plant DNA/RNA Kit.

Sample Preparation

- 1. Clean the samples as thoroughly as possible, taking care not to damage them.
- 2. Completely lyophilize the samples.
- 3. Weigh 25 mg of the lyophilized leafy gametophyte of *Polytrichum commune* or thallus of *Marchantia polymorpha*, and place it in a 2 ml microcentrifuge tube along with one 4 mm stainless bead.
- 4. Grind the sample into a fine powder using a TissueLyser II (or an equivalent bead-beater) at a frequency of 30 Hz for 1 min.
- 5. Add 800 μl of Buffer SQ1 and vortex vigorously for 30 s.
- 6. After briefly spinning down the sample, incubate it in a thermomixer at 1,500 rpm for 30 min at 60 °C.
- 7. Centrifuge at 13,000 rpm for 10 min at 4 °C.
- 8. Carefully transfer 300 µl of the supernatant to the 1st (7th) well of the cartridge, avoiding the fat and debris layer.
- 9. Add 10 μ l of RNase A Solution (20 mg/ml) to the 3rd (9th) well of the cartridge.

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Results

Sample (n=3)	Polytrichum commune		Marchantia polymorpha	
Sample (II-S)	Male (CV)	Female (CV)	Male (CV)	Female (CV)
Yield (µg)	34.0 (0.18)	18.4 (0.16)	18.4 (0.06)	23.0 (0.09)
A ₂₆₀ /A ₂₈₀	2.06 (0.01)	2.03 (0.01)	2.12 (0.00)	2.12 (0.00)
A ₂₆₀ /A ₂₃₀	2.01 (0.04)	1.88 (0.03)	2.16 (0.01)	2.22 (0.01)

Table 1. Evaluation of DNA yield and purity from bryophyte samples

The AllEx® Plant DNA/RNA Kit (n=3) was applied to extract genomic DNA from male and female samples of two bryophyte species using the AllEx®64 Automated Nucleic Acid Extraction System. The yield and purity of the extracted nucleic acids were measured using a NanoDrop™ 2000 spectrophotometer.

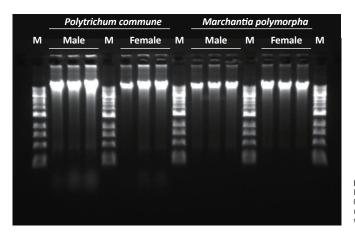


Figure 1. Agarose gel electrophoresis results of extracted genomic DNA from bryophyte samples Each extracted genomic DNA sample was subjected to electrophoresis on a 1% agarose gel with 0.5X TBE buffer at 150 V for 20 min. The sizes of the separated DNA fragments were confirmed using the GENESTA[™] 1 kb DNA Ladder (GA-100), and the electrophoresis results were analyzed with the SmartView Pro 1100 Imager System (UVCI-1100).

Conclusion

- Extraction of high-yield and high-quality nucleic acids from bryophytes is achievable using the AllEx[®]64 Automated Nucleic Acid Extraction System and its plant-specific extraction kit, the AllEx[®] Plant DNA/RNA Kit.
- The sample must be thoroughly pulverized. If it is not completely pulverized and forms clumps during the lysis step with Buffer SQ1, lysis efficiency may decrease. In such cases, vigorous vortexing can help disperse the clumps, resolving the issue and improving the lysis reaction.
- An excessively long incubation time can actually reduce the yield; therefore, the reaction time using the thermomixer should be strictly followed as 30 min, as specified in the protocol.

Ordering Information

Cat. No.	Product	Size
AEX064	AllEx [®] 64 Automated Nucleic Acid Extraction System	1 Unit
937-048	AllEx [®] Plant DNA/RNA Kit (Single Type)	48T
937-096	AllEx [®] Plant DNA/RNA Kit (Plate Type)	96T