iNtRON Biotechnology

AutoXT Plant DNA Kit (W/I)

For genomic DNA Extraction from various Plant tissues

Ver. INT-IFU-17604 (W/I)-R00

Cat. No

17604-96 (W); Well-plate type
17604-48 (I); Individual type

CONTENTS

INTRODUCTION

1. The AutoXT Plant DNA Kit is used in conjunction with the Miracle-AutoXT Nucleic Acid Extraction System (IMC-NC15PLUS) to purify genomic DNA from plant tissues such as leaves, stems, and seeds. This kit is designed for efficient and high-throughput DNA extraction, providing consistent and reliable results.

2. One of the key advantages of AutoXT Plant DNA Kit is its streamlined workflow-allowing for complete DNA extraction within 30 minutes, from sample preparation to final elution. Unlike many traditional plant DNA extraction methods, this protocol does not require additional high-temperature incubation steps, making it both convenient and time-efficient.

3. The purified DNA is of high quality, free from common plant-derived inhibitors such as polysaccharides and polyphenols, making it suitable for downstream applications such as PCR, qPCR, and next-generation sequencing (NGS). The Miracle-AutoXT Nucleic Acid Extraction System can process up to 32 samples simultaneously, maximizing efficiency in high-throughput settings.

PRODUCT COMPONENTS

Well plate type (W)

Contents	Unit	Q'ty
Prefilled Plates	16 test/Plate	6
Plunger Tips	ea	12
Pre-buffer[A]	40 mL/bottle	1
Pre-Columns[B]	48 ea/unit	2
RNase A (Lyophilized)[C]	3 mg/vial	4

Individual type (I)

Contents	Unit	Q'ty
Prefilled Cartridges	1 test/Cartridges	48
Plunger Tips	ea	12
Pre-buffer[A]	20 mL/bottle	1
Pre-Columns[B]	48 ea/unit	1
RNase A (Lyophilized)[C]	3 mg/vial	2

A. This Buffer contains chaotropic salt.
B. Inserted into a 2.0 ml collection tube (not provided)
C. The lyophilized RNase A can be stored at room temperature (15–25°C) until the kit's expiration date. It should be dissolved in distilled water (D.W.) and immediately stored at -20°C after dissolution. The solution is stable at -20°C for up to 24 months and can withstand 20 freeze-thaw cycle.

STORAGE CONDITIONS

AutoXT Plant DNA Kit should be stored dry, at room temperature (15–25°C). Under these conditions, AutoXT Plant DNA Kit can be stored for up to 24 months without showing any reduction in performance. The lyophilized RNase A can be stored at room temperature (15–25°C) until the kit's expiration date without affecting activity. The lyophilized RNase A can only be dissolved in D.W.; dissolved RNase A should be immediately stored at -20°C. The solution is stable at -20°C for up to 24 months and 20 times frozen-thawing until the kit's expiration date.

ADDITIONAL REQUIRED EQUIPMENT

AutoXT Plant DNA Kit provides all reagents for extracting DNA, including RNase A. However, be prepared some equipment and reagents as follows for a fast and easy extraction.

- Miracle-AutoXT Automated Nucleic Acid Extraction System (IMC-NC15PLUS)
- Pipettes and pipette tips
- Vortex mixer
- Microcentrifuge with rotor for 2.0 ml tubes
- Microcentrifuge tubes (1.5 ml)
- Liquid nitrogen
- Other general lab equipment

SAFETY INFORMATION

The reagent Cartridges or Plates contain ethanol which is flammable. Guanidine thiocyanate and Guanidine hydrochloride (which are components of the Binding Buffer and Washing Buffer 1) are harmful and irritants.
Always wear protective gear during handling chemical materials and the test should be handled by professionally trained person.

DO NOT add bleach or acidic solutions directly to the sample preparation waste.

PRODUCT WARRANTY AND SATISFACTION GUARANTEE

All products undergo extensive quality control test and are warranted to perform as described when used correctly. Satisfaction guarantee is conditional upon the customer providing full details of the problem to iNtRON within 60 days of purchase, and returning the product to iNtRON for examination.

CONSIDERATION BEFORE USE

1. Lyophilized RNase A

Note : Dissolve the RNase A in 0.3 ml of pure D.W.

2. Centrifugation : All centrifugation steps are carried out at RT (15 - 25°C).

NOTICE

1. For research purpose only.

2. Always wear protective gear during handling chemical materials and the test should be handled by professionally trained person.

3. Be careful and prevent the contamination and direct contact from the test samples.

4. Surface of workspace and pipette should be regularly sterilized by 10% bleach solution.

5. All the waste should be sterilized before discarding.

SAMPLE PREPATION

Recommended Volume of Starting Materials according to Plant samples

Amounts of starting material for AutoXT Plant DNA Kit procedures

1. Determine the appropriate amount (50 mg-100 mg) of plant material. Do not use more than 100 mg.

Note : Weighing plant tissue is the most accurate way to determine the amount.

2. Transfer the plant sample to a mortar, add liquid nitrogen, and thoroughly grind it into a fine powder using a pestle.

3. Transfer the sample into a 1.5 mL micro tube and add 300 µL of Pre-Buffer. Vortex vigorously.

Note : Ensure the sample is finely ground or homogenized before adding Lysis buffer to maximize DNA extraction efficiency.

4. Place the Pre-column into a 2 ml collection tube, then pipette the lysate directly into the column and centrifuge for 1 min at 13,000 rpm.

Note : After centrifugation, check the collection tube to ensure all the lysate has passed through. If residue remains in the column, an additional short spin may be necessary.

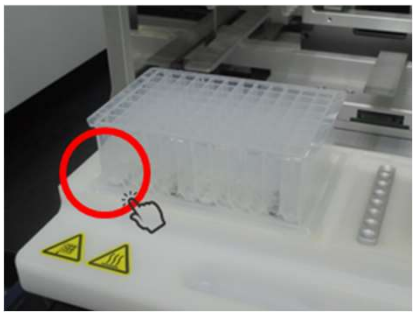
5. Transfer the 250 µL of flow-through into a new 1.5 mL microcentrifuge tube.

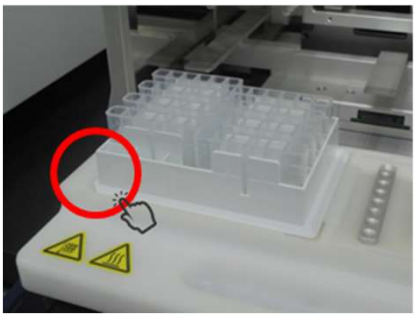
PROTOCOL

1. Add 10 µL of RNase A solution to to each of the wells 2 and 8.
Note : Ensure that the RNase A solution is fully thawed and mixed before use to maintain enzyme activity.

2. Add 200 µL of samples from [sample preparation Step] to each of the wells 1 and 7.

3. Insert Prefilled Well-Plate or Cartridge Rack combined with Prefilled Cartridge on Heating Tray.
Note : Make sure the position of the diagonally cut edge of plat forward on the Heating Tray.

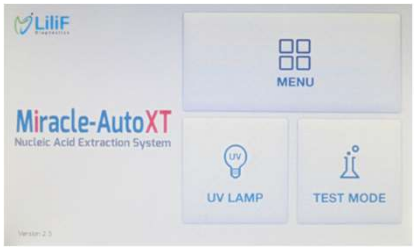


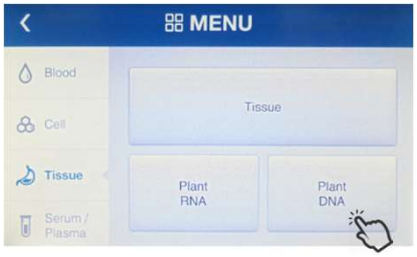


4. Close the front door and ready to start.

5. Press the 'menu / Tissue' button on the touch display of the Miracle-AutoXT Nucleic Acid Extraction System to select the extraction type.

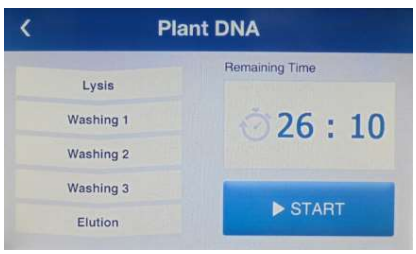
6. Select 'Plant DNA' icon for plant DNA extraction as shown figure below.





7. Press the 'Start' button to perform the extraction.

8. After completion of device working, transfer the 60~80 µL of Elution fraction (well position 6) to a new 1.5 ml Microtube.



TROUBLESHOOTING GUIDE

Problem	Possible Cause and Recommendation
Low DNA yield	- Inadequate tissue disruption. ensure proper grinding or homogenization before adding lysis buffer. Using liquid nitrogen, a bead beater, or a mechanical homogenizer can help finely disrupt tough plant tissues. Insufficient grinding may leave intact cells, reducing DNA release and affecting downstream applications. - Tissue has low DNA. content. Some plant tissues have inherently low DNA content. For example, stems, woody tissues, and some mature leaves contain fewer nucleated cells compared to young leaves or seeds. Stems and vascular tissues are often composed of lignified cells or dead cells, which naturally have lower DNA yields. - Too much starting material. Using an excessive amount of starting material can overwhelm the lysis buffer, leading to incomplete cell disruption and reduced DNA yield. Overloaded samples may result in poor buffer penetration, leaving intact cells and causing inefficient DNA extraction. - Sample material not stored properly. Whenever possible, use fresh material. If this is not possible, flash freeze the samples in liquid nitrogen. Samples should always be kept at -70°C. Never allow tissues to thaw before addition of Lysis buffer. Perform disruption of samples in liquid nitrogen. - The Miracle-AutoXT Nucleic Acid Extraction System Instrument was set for the wrong method. Ensure that the correct method is chosen in Plant DNA Mode. - Check that Plunger Tips were added to the plates/cartridges. Ensure that all plates/cartridges are snapped into the rack properly before processing.
Poor amplification	- Paramagnetic particle carryover may cause interference in amplification reaction. Remove particles in Elution Tube by centrifugation.
Cross-contamination	- Avoid splashing when adding lysates to plates/cartridges. Plates/Cartridges may be removed from the rack for sample addition to minimize contamination of adjacent paltes/cartridges.se fresh plastic wares for each sample to prevent sample-to sample contamination.
Plant method not an option on the instrument	- For the Miracle-AutoXT Nucleic Acid Extraction System Instrument, verify that the instrument is in Engineer mode. Verify that the instrument has firmware which includes the Plant DNA method.

EXPERIMENTAL INFORMATIONS

DNA extraction efficiency from different types of plant samples

The iNtRON's AutoXT Plant DNA Kit gave improved DNA yield compare with competitor from various plant samples.

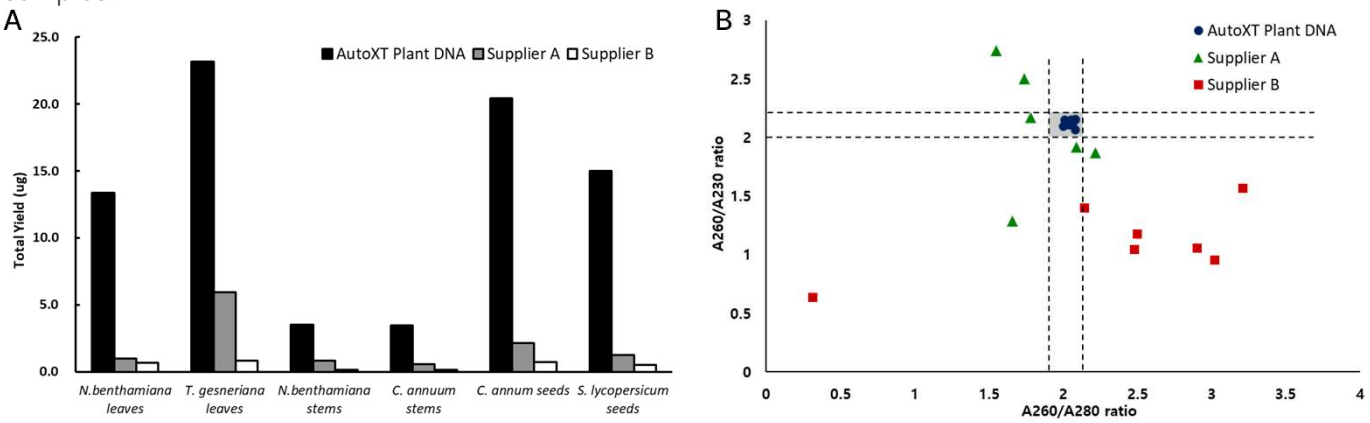


Fig. 1. Genomic DNA yield and purity were measured various plant samples.
(A) Total DNA yield (µg) extracted from various plant samples using the AutoXT Plant DNA Kit (black bars), Supplier A (gray bars), and Supplier B (white bars). The AutoXT Plant DNA Kit consistently yields the highest DNA amounts across all tested plant tissues, including leaves, stems, and seeds.
(B) Scatter plot showing DNA purity, measured as A260/A280 vs. A260/A230 ratios, for samples extracted with competitor. The AutoXT Plant DNA Kit (blue dots) demonstrates high purity, clustering within the optimal range (A260/A280 ≈ 2.0, A260/A230 ≈ 2.0–2.2, shaded region), while competitor kits (Supplier A: green triangles, Supplier B: red squares) exhibit greater variability and lower purity values.

High quality of extracted DNA

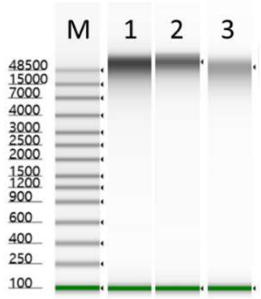


Fig. 2. Genomic DNA extracted using the AutoXT Plant DNA Kit was run on an Agilent 4200 TapeStation System and is displayed here as a gel electropherogram.
The electropherogram confirms the presence of intact, high-molecular-weight genomic DNA bands with minimal degradation.
Lane M, 35 bp lower marker; Lane 1, *T. gesneriana* leaves; Lane 2, *C. annuum* seeds; Lane 3, *S. lycopersicum* seeds

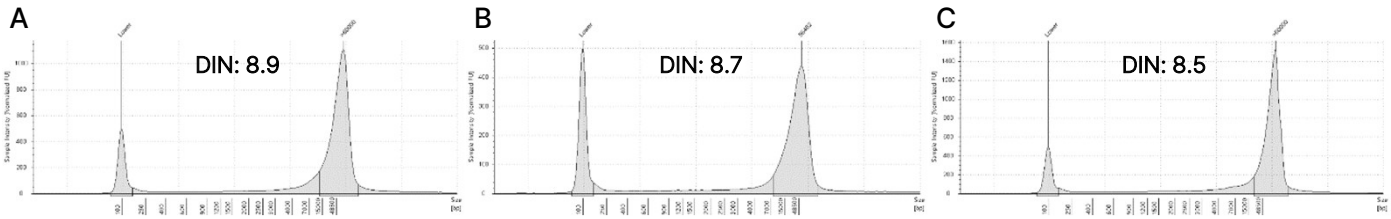


Fig. 3. Electropherogram profiles of genomic DNA extracted from plant samples using AutoXT Plant DNA Kit. The DNA Integrity Number (DIN) values were assessed to evaluate the integrity of the extracted DNA.
These results confirm that the AutoXT Plant DNA Kit effectively preserves DNA quality, making it suitable for downstream applications such as PCR, qPCR, and next-generation sequencing (NGS).
A, *T. gesneriana* leaves; B, *C. annuum* seeds; C, *S. lycopersicum* seeds.

EXPERIMENTAL INFORMATIONS

Determination of yield and purity data of various plant samples

The genomic DNA extraction results using AutoXT Plant DNA Kit were shown high quality and quantity of DNA collected from 50 mg of various plant tissue samples.

Type	Sample	Lane	DNA yield (μg)	A260/280
Leaves	*N. benthamiana*	1	13.3	2.08
	N. tabacum	2	7.1	2.05
	C. unshiu	3	11.5	2.06
	C. melo	4	16.6	2.11
	C. pepo	5	15.9	2.11
	C. quinoa	6	10.4	2.07
	C. annuum	7	9.2	2.12
	C. amaranticolor	8	19.0	2.07
	D. stramonium	9	8.6	2.05
	S. lycopersicum	10	9.1	2.06
	O. Sativa	11	13.6	2.06
	T. gesneriana	12	23.2	2.07
Stems	*N. benthamiana*	13	3.5	2.10
	N. tabacum	14	5.4	2.01
	C. annuum	15	3.4	2.04
	C. melo	16	2.9	2.08
	C. amaranticolor	17	5.9	2.05
	S. lycopersicum	18	3.7	2.05
Seeds	*N. benthamiana*	19	24.5	2.07
	C. annuum	20	20.4	2.08
	C. sativus	21	23.3	2.09
	A. thaliana	22	19.1	2.09
	S. lycopersicum	23	15.0	2.06
	G. max	24	32.3	2.14

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