

**VAMNE Magnetic Universal Plant
Total RNA Kit (Prepackaged)**

RMA4103



Instruction for Use

Version 26.1

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For Research Use Only. Not for use in diagnostic procedures.

01/Product Description

VAMNE Magnetic Universal Plant Total RNA Kit (Prepackaged) is designed for the extraction of high-purity RNA from various plant tissues, achieving high-throughput processing of samples. The kit is based on superparamagnetic particle purification technology with an optimized reagent system, which can specifically adsorb nucleic acids and effectively remove various impurities. DNase I can efficiently eliminate DNA. The fully automated workflow is simple, rapid, safe and efficient. The obtained RNA can be directly used for downstream experiments such as RT-PCR, qRT-PCR, Northern Blot, Dot Blot, *in vitro* translation, and high-throughput sequencing.

02/Components

Components		RMA4103-01 (1 × 96 T)
BOX 1	Binding (Plate 1)	1
	Beads (Plate 2)	1
	Wash 1 (Plate 3)	1
	Wash 2 (Plate 4)	1
	Wash 3 (Plate 5)	1
	Elution (Plate 6)	1
	Lysis Buffer PLA	80 ml
	Buffer RDD	5 ml
	☐ Elution Buffer	1 ml
BOX 2	Proteinase K	2.5 ml
	 Buffer TP	3 × 1.2 ml
	 DNase I	320 μl

03/Storage

BOX 1: Store at 15 ~ 25°C and ship at room temperature.

BOX 2: Store at -30 ~ -15°C and ship at ≤0°C.

04/Applicable Instruments

For use with the fully automatic nucleic acids extraction instruments (Vazyme #VNP-96P) and equivalent instruments (Heating positions: Positions 1 and 6).

▲ For automated instruments of other brands or models, compatible consumables and operating programs must be adjusted accordingly.

05/Applications

Plant leaves and fungi: 20 - 100 mg;

Polysaccharide-rich tubers, roots and seeds: 20 - 50 mg;

Fruits: 50 - 200 mg.

06/Self-prepared Materials

Seed Lysis Buffer (Vazyme #DMA4101-C2) (Optional).

07/Notes

1. The key to successful RNA extraction is to prevent RNase contamination. RNases are ubiquitous and extremely stable in the environment. Even trace amounts of RNase can rapidly degrade RNA. Therefore, please follow standard RNA extraction precautions, including wearing masks and disposable sterilized gloves, operating in a dedicated and clean area, and using RNase-free laboratory consumables.
2. The automatic nucleic acids extraction instruments should be disinfected with the UV light for 30 min before and after use.
3. Traces of magnetic beads may remain in the eluate after extraction. Avoid aspirating the magnetic beads. If magnetic beads are accidentally aspirated, repeat magnetic adsorption once more using a magnetic rack.
4. Properly dispose of all samples and reagents, thoroughly wipe down and disinfect all work surfaces with 75% ethanol.

08/Experiment Process

08-1/Reagents Preparation

Take out the prepackaged plate from the kit. Invert the plate several times to fully resuspend the magnetic beads. Gently flick the plate to concentrate the reagents and beads at the bottom of the well. Before use, ensure the plate orientation and carefully remove the aluminum sealing film.

▲ Avoid vibration when tearing off the sealing foil to prevent liquid from spilling.

08-2/Sample Pretreatment

1. Sample grinding: Rapidly transfer fresh or frozen plant tissue stored at -85 ~ -65°C into a liquid nitrogen-precooled mortar and grind thoroughly. Continuously add liquid nitrogen to maintain the tissue at a low temperature until it is ground into a fine powder with no obvious particles. Alternatively, rapidly grind the tissue into a powder at low temperature using a fully automated tissue homogenizer. Recommended parameters are provided in the table below.

Sample type	Parameters settings
Plant leaves, flowers, fruits, fungi, etc.	40 - 60 Hz 45 sec, 1 cycle
Plant polysaccharide-rich tubers, roots, etc.	40 - 60 Hz 45 sec, 2 cycles
Plant seeds such as soybeans, wheat seeds, etc.	60 Hz 45 sec, 3 cycles

▲ The grinding parameters can be adjusted according to the difficulty of disrupting sample. Laboratory-standard grinding parameters may also be used as appropriate.

▲ All grinding accessories should be pre-cooled with liquid nitrogen prior to use, and re-precooled between successive grinding cycles.

2. Sample Pre-processing Solution preparation: Add 30 µl Buffer TP and 20 µl Proteinase K to each 800 µl of Lysis Buffer PLA and mix well. Transfer the ground tissue into a 1.5 ml centrifuge tube. Immediately add 850 µl Pre-processing Solution. Vigorously vortex for 30 - 60 sec to thoroughly mix the sample and Pre-processing Solution until no obvious large tissue clumps remain, then incubate at room temperature for 5 min. Centrifuge at 12,000 rpm (~13,400 × g) for 5 min at room temperature. Transfer 700 µl of the supernatant to the Binding Plate Binding (Plate 1).

▲ Pre-processing Solution needs to be prepared freshly and used immediately. It is recommended to prepare Pre-processing Solution for N + 1 reactions.

▲ For samples with low RNA content, the input amount can be appropriately increased. Due to the diversity of plant samples, the input amount can be adjusted based on specific experimental requirements and experience.

▲ For certain seed or polysaccharide-rich samples, a transparent gel-like layer may appear between the pellet and the supernatant after centrifugation. Accidentally aspirating this layer can affect extraction efficiency. To ensure optimal results, the volume of the supernatant can be appropriately reduced. For seed samples (e.g., wheat seeds, rice seeds) ≥30 mg, Seed Lysis Buffer (Vazyme #DMA4101-C2) is recommended. Contact sales representatives for acquisition. Contact sales representatives for acquisition.

08-3/Aliquoting DNase I

1. DNase I Working Solution Preparation: For each reaction, mix 47 µl Buffer RDD with 3 µl DNase I by gentle pipetting.

2. Add the prepared DNase I Working Solution to the wells of Beads Plate (Plate 2), dispensing 50 µl per well.

▲ It is recommended to prepare DNase I Working Solution for N + 2 reactions.

▲ DNase I Working Solution needs to be prepared freshly and used immediately. Make sure to add the DNase I Working Solution directly into the liquid; adding it onto the well wall may reduce the efficiency of genomic DNA removal.

08-4/Operation on the automatic instrument

1. Place the 96-deep-well plates into the Automatic nucleic acids extraction instrument in the following order: Binding (Plate 1), Beads (Plate 2), Wash 1 (Plate 3), Wash 2 (Plate 4), Wash 3 (Plate 5) and Elution (Plate 6). **Position the deep-well plates with the notched corner toward the inner right front of the instrument.** Place the magnetic rod sleeves into the Beads Plate (Plate 2) and ensure they are properly seated.

2. Edit the program as below (or select the corresponding pre-imported program) to perform automated extraction.

The Program for Vazyme VNP-96P (RMA4103)

Step	Plate Position	Name	Mixing Time (min)	Adsorption Time (sec)	Waiting Time (min)	Volume (μl)	Mixing Speed	Temperature (°C)	Mixing Position	Mixing Amplitude	Adsorption Position	Adsorption Speed
1	2	Beads	0.1	0	0	300	8	-	10	80	0	10
2	1	Lysis	5	0	0	900	8	58	10	80	0	10
3	2	Beads	0.5	30	0	300	8	-	10	80	0	10
4	1	Lysis	2	60	0	900	8	58	10	80	0	10
5	3	Wash 1	1	30	0	700	8	-	10	80	0	10
6	2	DNase I	10	30	0	300	8	-	10	80	0	10
7	3	Wash 1	1	30	0	700	8	-	10	80	0	10
8	4	Wash 2	1	30	0	700	8	-	10	80	0	10
9	5	Wash 3	1	30	5	700	8	-	10	80	0	10
10	6	Elution	2.5	60	0	70	10	75	10	80	0	10
11	2	Beads	0.1	0	0	300	8	-	10	80	0	10

Other settings (in the Option menu):

Temperature control (TC) setting (TC Type: Start with TC); **Adsorption setting (Adsorption type: three-stage adsorption)**;
Drying setting (Drying position: Above the kit; Drying fan: OFF)

- After the automated extraction, promptly transfer the eluates from Elution plate (note the valid wells) into clean RNase-free centrifuge tubes. The extracted product can be stored at -85 ~ -65°C for long-term storage, and -30 ~ -15°C for short-term storage.



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