

ONEMag™ Rapid Plant RNA Extraction Kit

(ONE-PLRNA-50)

Principle:

The **ONEMag™ Rapid Plant RNA Extraction Kit** is based on magnetic bead-based purification technology for rapid and efficient isolation of high-quality RNA from different Plant samples.

The lysis conditions are optimized to inactivate RNases and minimize degradation of the released RNA. Plant-derived contaminants such as polysaccharides, polyphenols, pigments, proteins, and other secondary metabolites are removed during the separation of cellular debris and purification steps.

Following lysis and separation of cellular debris, the released RNA is subjected to optimized chaotropic salt conditions that promote selective binding of RNA to silica-coated magnetic beads. The magnetic beads are separated using a magnetic field and washed sequentially to remove residual proteins, polysaccharides, phenolic compounds, salts, and other plant-derived inhibitors. A brief air-drying step ensures complete removal of wash buffer residues. The purified RNA is subsequently eluted under low-salt conditions.

The isolated RNA is suitable for downstream applications including RT-PCR, RT-qPCR, gene expression analysis, transcriptome studies, RNA sequencing (RNA-Seq), and other RNA-based molecular applications.

Kit Contents:

Components	Quantity (50 Reactions)
PRL Buffer	30 mL
RBB Buffer	30 mL
RCB Beads	3 mL
RWB Buffer	15 mL
REB Buffer	4 mL

Material's/Equipment's Required but not Supplied:

- Ethanol (96–100%), Molecular Grade
- Micropipettes and sterile tips
- 1.5 mL Microcentrifuge Tubes
- Heating block or Water bath
- Vortex Mixer
- Magnetic Stand for 1.5/2.0 mL tubes
- DNase I

Storage Conditions:

All components of the **ONEMag™ Rapid Plant RNA Extraction Kit** should be stored at room temperature (~25°C). The kit is stable until the expiration date printed on the product label when stored under recommended conditions.

Preparation of Buffers

Preparation of RWB Buffer

RWB Buffer is supplied as a **concentrate** and must be diluted with ethanol before use.

Component	Volume for 50 Reactions
RWB Buffer (Concentrate)	15 mL
Ethanol (96-100%)	45 mL
Final Volume	60 mL

Mix thoroughly and store at room temperature.

Protocol:

1. Weigh up to 50- 100 mg of sample and homogenize it accordingly.
2. Add 500 μ L PRL Buffer and incubate at 65°C for 10 minutes to ensure efficient mechanical lysis.
(Add 10 μ L of DNase I to the lysate and mix thoroughly. Incubate at 37 °C for 20 minutes. This step is recommended for efficient removal of genomic DNA and to improve the purity and quality of the extracted RNA).
3. Centrifuge at 5,000 rpm for 8 minutes and transfer 500 μ L of lysate to a new 1.5 mL tube.
4. Add 500 μ L of RBB Buffer and vortex well.
5. Add 20 μ L of RCB Bead and vortex thoroughly.
6. Incubate at room temperature for 5 minutes.
7. Place the tube on a magnetic stand and allow the beads to separate for 30 seconds.
8. Carefully discard the supernatant without disturbing the beads.
9. Add 500 μ L of RWB Buffer and vortex well.
10. Place the tube on the magnetic stand for 30 seconds.
11. Carefully discard the supernatant.
12. Add 500 μ L of RWB Buffer, vortex, place on the magnetic stand for 30 seconds, discard the supernatant, and air-dry the beads for 1 minute (do not over-dry).
13. Add 50 μ L of REB Buffer, vortex briefly, and incubate at room temperature for 5 minutes.

14. Place the tube on the magnetic stand for 1 minute, then transfer the eluted RNA to a new tube and store at -20°C .

Troubleshooting Guide:

Problem	Possible Cause	Recommended Solution
Low RNA yield	Incomplete mechanical or chemical lysis	Ensure the plant tissue is thoroughly homogenized and processed with the recommended lysis buffer. Vortex vigorously for the specified time and incubate at 65°C for 15 minutes.
	Insufficient sample homogenization	Homogenize the plant tissue thoroughly to ensure complete cell disruption and efficient RNA release.
	Loss of magnetic beads during supernatant removal	Place the tube fully on the magnetic stand and remove the supernatant carefully without disturbing the beads.
Poor RNA purity	Carryover of polysaccharides, polyphenols, or other plant-derived inhibitors	Perform all washing steps completely. If contamination persists, repeat the recommended wash step once more.
	Incorrect wash buffer preparation	Confirm that the wash buffer concentrate has been diluted with the correct volume of ethanol before use.
	Residual wash buffer or ethanol	Allow the beads to air-dry for approximately 1 minute before elution. Avoid excessive drying of the beads.
Beads do not separate properly	Magnetic stand not suitable	Use a magnetic stand compatible with the extraction tube size and ensure firm contact between the tube and magnetic surface.
	Insufficient separation time	Allow the beads to collect completely on the magnetic stand for at least 30 seconds before removing the supernatant.
Cloudy or viscous lysate	High polysaccharide, polyphenol, or other secondary metabolite content	Increase centrifugation time slightly and carefully transfer only the clear lysate for the subsequent binding step.
	Excess plant tissue input	Do not exceed the recommended sample input for the extraction procedure.
Low RT-PCR / RT-qPCR performance	Residual plant-derived inhibitors	Ensure complete washing and removal of residual ethanol. If inhibition persists, dilute the RNA appropriately before downstream application.

Problem	Possible Cause	Recommended Solution
	Elution step missed or incorrect	Add the recommended elution buffer directly to the beads, mix thoroughly, and incubate for the specified time before magnetic separation.
RNA degradation	RNase contamination	Use RNase-free tubes, tips, and reagents. Wear fresh gloves and avoid unnecessary handling of the RNA sample.
	Improper sample handling or storage	Process samples promptly and keep RNA on ice during handling. Store purified RNA at -80°C for long-term storage and avoid repeated freeze–thaw cycles.
Genomic DNA contamination	Insufficient DNase treatment	Perform the recommended DNase I treatment using $10\ \mu\text{L}$ DNase I and incubate at 37°C for 20 minutes.
Low RNA integrity	Delayed processing or RNase exposure	Minimize the time between tissue homogenization and maintain RNase-free conditions throughout the procedure.