

Mag Beads Plant RNA Extraction Kit

【Product Name】

Mag Beads Plant RNA Extraction Kit

【Package Specifications】

50T/box (Art.No. FP102-50); 100T/box (Art.No.FP102-100)

【Intended Use】

Suitable for extracting total RNA from 50 to 100 mg of plant samples. Also compatible with high-throughput workstations and other automated nucleic acid extraction devices. The obtained RNA can be directly used for downstream experiments such as Northern Blot and in vitro translation.

【Detection Principle】

RNA binds to the surface of Magbeads coated with silica under high salt and low pH conditions. After multiple washes to remove impurities such as proteins, RNA is eluted under low salt conditions, resulting in high-purity RNA.

【Main Components】

| Components | FP102-50 | FP102-100 |
|------------------|----------|-----------|
| Bead suspension | 1.1mL | 2×1.1mL |
| NP Columns | 50 | 100 |
| Buffer PST | 40mL | 80mL |
| Buffer W1A* | 30mL | 66mL |
| Buffer W2R* | 15mL | 30mL |
| RNase Free Water | 10mL | 20mL |

【User-provided Equipment and Reagents】

Equipment : Nucleic acid extraction instrument, 2.2 mL 96-well deep well plates (U-bottom), magnetic rack, vortex mixer, constant temperature oscillator, etc.

Reagents: Anhydrous ethanol, isopropanol.

【Storage Conditions and Shelf Life】

This product can be stored at room temperature (15-25°C) for 12 months.

【Precautions】

1. Before using Buffer W1A/W2R, dilute with absolute ethanol as indicated on the bottle label.
2. Avoid repeated freeze-thaw cycles of samples.
3. Magnetic beads should not be frozen, and the magnetic bead suspension should be thoroughly mixed before use.
4. Wear protective equipment before operation. In case of accidental skin or eye contact with reagents, immediately rinse with water for 5-10 minutes.
5. Before each use, check if the lysis solution has precipitated. If so, re-dissolve at 60°C.

【Manual Operation Steps using Centrifuge Tubes】

1. Sample Pretreatment

1) Take plant samples for liquid nitrogen grinding, weigh 50-100 mg of sample, add 600μL of Buffer PST, and vigorously vortex for 20-60 seconds to disperse the sample. Incubate at room temperature for 5 minutes.

2) Centrifuge at 13,000xg for 1 minute, transfer 550μL of supernatant to NP columns, centrifuge at 13,000 xg for 1 minute, and set aside.

2. **RNA Binding:** Add 20μL of magnetic bead suspension and 350μL of isopropanol to

the pretreated supernatant, mix thoroughly by vortexing. Incubate at room temperature for 5 minutes, invert several times during incubation.

3. **Magnetic Separation:** Place the centrifuge tube on the magnetic rack and let it stand for 30 seconds until the magnetic beads are completely adsorbed, carefully discard the liquid.

4. **Wash 1:** Remove the centrifuge tube from the magnetic rack, add 600 μ L of Buffer W1A (confirm ethanol addition), vortex for 1 minute. Place the centrifuge tube on the magnetic rack and let it stand for 30 seconds until the magnetic beads are completely adsorbed, carefully discard the liquid.

5. **Wash 2:** Remove the centrifuge tube from the magnetic rack, add 600 μ L of Buffer W1A (confirm ethanol addition), vortex for 1 minute. Place the centrifuge tube on the magnetic rack and let it stand for 30 seconds until the magnetic beads are completely adsorbed, carefully discard the liquid.

6. **Wash 3:** Remove the centrifuge tube from the magnetic rack, add 600 μ L of Buffer W2R (confirm ethanol addition), vortex for 1 minute. Place the centrifuge tube on the magnetic rack and let it stand for 30 seconds until the magnetic beads are completely adsorbed, carefully discard the liquid.

7. **Wash 4:** Repeat step 6 once.

8. **Magnetic Separation:** Briefly centrifuge, place the centrifuge tube on the magnetic rack, and let it stand for 10 seconds to remove residual liquid (this step is crucial to avoid ethanol residue that may inhibit subsequent experiments).

9. **De-alcoholization:** Place the centrifuge tube on the magnetic rack, open the lid, and air dry at room temperature for 5-10 minutes (excessive drying time may lead to difficulty in elution).

10. **Elution:** Remove the centrifuge tube, add 50-100 μ L of RNase Free Water, vortex thoroughly, and shake at 1500 rpm at 55 $^{\circ}$ C for 5 minutes (alternatively, use a 55 $^{\circ}$ C water bath and vortex mix 3-4 times during the incubation).

11. **Nucleic Acid Transfer:** Place the centrifuge tube on the magnetic rack, let it stand for 2 minutes to allow magnetic bead adsorption, then transfer the RNA solution to a new centrifuge tube and store at -80 $^{\circ}$ C.

【Operation Steps for QP-AUT-16/32 Channel Nucleic Acid Extractor】

1. Sample Preparation:

Add the specified amounts into each corresponding well of the 96-well plate as shown in the table below, enabling the simultaneous processing of 16/32 samples.

| Position | 1/7 | 2/8 | 3/9 | 4/10 | 5/11 | 6/12 |
|----------|---|-----------------------------|-----------------------------|-----------------------------|-----------------------------|---|
| Reagent | Binding Buffer (20 μ L) +Isopropanol (350 μ L) | Buffer W1A (600 μ L) | Buffer W1A (600 μ L) | Buffer W2R (600 μ L) | Buffer W2R (600 μ L) | RNase Free Water (50-100 μ L) |

2. Sample Preparation:

1) Take plant samples for liquid nitrogen grinding, weigh 50-100 mg of sample, add 600 μ L of Buffer PST, vortex vigorously for 20-60 seconds to disperse the sample, and let it stand at room temperature for 5 minutes.

2) Centrifuge at 13,000 x g for 1 minute, transfer 550 μ L of supernatant to NP Columns, centrifuge again at 13,000 x g for 1 minute, and set aside.

3. Automated Extraction:

Transfer the processed supernatant to wells 1/7 of a 96-deep well plate, then sequentially place the prepared 96-well sample plate into the nucleic acid extractor. Insert

the magnetic rod sleeve, open the instrument's operating program, select the corresponding program, click "run", and start the extraction.

4. Nucleic Acid Transfer:

After the instrument run is complete, transfer the eluate from wells 6/12 of the 96-deep well plate to clean, nuclease-free centrifuge tubes.

The parameters for the 32-channel nucleic acid extractor (QP-AUT-32) program are set as follows:

| Step | Site | Name | Waiting time(min) | Mixing time (min) | Suction time(sec) | Volume (μL) | Mixing velocity | Temperature |
|---------|------|---------------|-------------------|-------------------|-------------------|-------------|-----------------|-------------|
| 1 | 1 | Lysis Binding | 0 | 5 | 60 | 1000 | 2 | OFF |
| 2 | 2 | Washing 1 | 0 | 2 | 60 | 600 | 3 | OFF |
| 3 | 3 | Combine | 0 | 15 | 0 | 80 | 3 | OFF |
| Suspend | | | | | | | | |
| 4 | 3 | Combine | 0 | 5 | 60 | 550 | 3 | OFF |
| 5 | 4 | Washing2 | 0 | 1 | 60 | 600 | 3 | OFF |
| 6 | 5 | Washing3 | 0 | 1 | 60 | 600 | 3 | OFF |
| 7 | 6 | Elution | 2 | 5 | 60 | 100 | 3 | 60°C |
| 8 | 5 | Abandon beads | 0 | 1 | 0 | 600 | 2 | OFF |

【Operation Steps of the 96-channel Nucleic Acid Extractor】

1. Sample Loading:

Prepare samples by adding the specified amounts into each well of the 96-well plates according to the table below. This allows simultaneous processing of 96 samples.

| | | | | | | |
|----------|---|---|---|---|---|---|
| Position | 1 | 2 | 3 | 4 | 5 | 6 |
|----------|---|---|---|---|---|---|

| Reagent | Bead Suspension (20μL) +isopropanol (350μL) | Buffer W1A (600μL) | Buffer W1A (600μL) | Buffer W2R (600μL) | Buffer W2R (600μL) | RNase Free Water (50-100μL) |
|---------|--|--------------------|--------------------|--------------------|--------------------|-----------------------------|
|---------|--|--------------------|--------------------|--------------------|--------------------|-----------------------------|

2. Sample Preparation:

1) Take plant samples for liquid nitrogen grinding, weigh 50-100 mg of sample, add 600μL of Buffer PST, vortex vigorously for 20-60 seconds to disperse the sample, and let it stand at room temperature for 5 minutes.

2) Centrifuge at 13,000xg for 1 minute, transfer 550μL of supernatant to NP Columns, centrifuge again at 13,000xg for 1 minute, and set aside.

3. Automated Extraction:

Transfer the processed supernatant to well 1 of a 96-deep well plate, then sequentially place the prepared 96-well sample plate into the nucleic acid extractor. Insert the magnetic rod sleeve, open the instrument's operating program, select the corresponding program, click "run", and start the extraction.

4. Nucleic Acid Transfer:

After the instrument run is complete, either seal or transfer the eluate from well 6 directly to clean, nuclease-free centrifuge tubes and store at -20°C for later use.

The parameters for the 96-channel nucleic acid extractor (QP-AUT-96) program are set as follows

| Procedure | Step 1 | Step 2 | Step 3 | Step 4 | Step 5 | Step 6 | Step 7 | Step 8 |
|--------------|----------|----------|----------|----------|----------|----------|----------|----------|
| Station | 1 | 2 | 3 | 3 | 4 | 5 | 6 | 5 |
| Waiting time | 00:00:00 | 00:00:00 | 00:00:00 | 00:00:00 | 00:00:00 | 00:00:00 | 00:02:00 | 00:00:00 |

| | | | | | | | | |
|--------------|--------------|-------------|------------|-------------|-------------|-------------|-------------|-------------|
| Mixed model | 2 | 3 | 3 | 3 | 3 | 3 | 3 | 2 |
| Mixing time | 00:05:00 | 00:02:00 | 00:15:00 | 00:05:00 | 00:01:00 | 00:01:00 | 00:05:00 | 00:00:30 |
| Suspend | No | No | No | No | No | No | No | No |
| Suction time | 00:01:00 | 00:01:00 | 00:00:00 | 00:01:00 | 00:01:00 | 00:01:00 | 00:01:00 | 00:00:00 |
| Volume | 1000 μ L | 600 μ L | 80 μ L | 550 μ L | 600 μ L | 600 μ L | 100 μ L | 600 μ L |
| Temperature | — | — | — | — | — | — | 60°C | — |

【Basic Information】

Version Number: 1.1

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Nanjing Rebeads Biotech Co., Ltd.

Address: 9th Floor, Building D, No. 606 Ningliu Road, Chemical Industry Park, Jiangbei New District, Nanjing

Postal Code: 210000

Phone: 025-58069660

Email: order@rebeads.com.cn