Mag Beads Blood RNA Extraction Kit

【Product Name】

Mag Beads Blood RNA Extraction Kit

【Package Specifications】

50T/box（Art.No.FP103-50）；100T/box（Art.No.FP103-100）

【Intended Use】

Suitable for extracting RNA from fresh blood samples. Also compatible with high-throughput workstations and other nucleic acid automated extraction devices. The resulting 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 silicon 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	FP103-50	FP103-100
Bead Suspension	1.1 mL	2×1.1 mL
ONRol Reagent	55 mL	105 mL
Buffer RBC	45 mL	90 mL
Buffer W1A*	22 mL	44mL
Buffer W2R*	15 mL	30mL
RNase Free Water	10 mL	20mL

【Equipment and Reagents】

1．Equipment: Nucleic acid extractor, 2.2mL 96-well deep well plates (U-bottom), magnetic rod covers, centrifuge tubes, magnetic racks, vortex oscillators, pipettes, constant temperature oscillators, etc.

2．Reagents: Absolute ethanol, chloroform.

【Storage Conditions and Shelf Life】

Except for ONRol Reagent and Buffer RBC, this product can be stored at room temperature (15~25°C) for 12 months. Since RNase-Free Water does not contain antimicrobial agents, storing or operating at room temperature may introduce bacterial or fungal contamination. It is recommended to aliquot and store at 2~8°C to minimize contamination.

【Precautions】

1．Before using Buffer W1A/W2R, dilute with absolute ethanol as indicated on the bottle label.

2．Avoid repeated freeze-thaw cycles for samples.

3．Do not freeze the magnetic beads. Thoroughly mix the magnetic bead suspension before use.

4．Wear protective equipment before handling. In case of accidental splashing onto the skin or eyes, immediately rinse with water for 5-10 minutes.

5．Check for any precipitation in the lysis buffer before each use. If present, dissolve again at 60°C.

【Manual Operation Steps for Centrifuge Tube】

1．Sample Handling and Lysis

1) In a 15mL centrifuge tube, add 1 volume of blood (≤1.5mL) and 5 volumes of

Buffer RBC (dilute the reagent as indicated on the label, recommend preparing fresh), invert and mix 5-10 times.

2) Place on ice for 10-15 minutes, invert and mix twice during this period. During incubation, the blood will change from misty to clear solution, indicating lysis of red blood cells. Centrifuge at 4°C, 500 x g for 10 minutes, carefully discard the supernatant.

3) Add 2 volumes of Buffer RBC to the blood sample (dilute the reagent as indicated on the label, recommend preparing fresh), briefly vortex to resuspend cells.

Note: If the initial blood volume is 1.5mL, add 3 mL of Buffer RBC.

4) Centrifuge at 4°C, 500xg for 10 minutes, carefully discard the supernatant.

Note: Try to remove residual solution, residual liquid should not exceed 100μL. Collected white blood cell precipitate can be directly stored at -80°C.

5) Immediately add 1mL of ONRol Reagent to the white blood cell precipitate. Vortex to resuspend cells.

Note: It is recommended to pipette up and down 5-10times to homogenize the sample, which helps to increase yield and stability.

6) After thorough vortexing, transfer the sample to a new 2mL centrifuge tube, and incubate at room temperature for 5-10minutes for complete cell lysis.

7) Add 200μL of chloroform to the lysis solution. Vigorously shake by hand for 15s; incubate at room temperature for 3 minutes.

Note: Using vortex instead of shaking will result in more genomic DNA contamination. Chloroform must be added in proportion, excessive chloroform will force DNA and proteins back into the aqueous phase, leading to decreased RNA purity.

8) Centrifuge at 4°C, 12,000xg for 15minutes, transfer the supernatant to a new centrifuge tube, and set aside.

2. Binding:

Add 20μL of magnetic bead suspension and 350μL of absolute ethanol to the lysate. Vortex thoroughly. Incubate at room temperature for 5 minutes, swirling occasionally.

3. Magnetic Separation:

Place the centrifuge tube on a magnetic rack and let it stand for 30 seconds until the magnetic beads are fully adsorbed, then carefully remove the liquid.

4. Wash 1:

Remove the centrifuge tube from the magnetic rack, add 600μL of Buffer W1A, vortex for 1 minute. Place the centrifuge tube on the magnetic rack and let it stand for 30 seconds until the magnetic beads are fully adsorbed, then carefully remove the liquid.

5. Wash 2:

Remove the centrifuge tube from the magnetic rack, add 600μL of Buffer W2R, vortex for 1 minute. Place the centrifuge tube on the magnetic rack and let it stand for 30 seconds until the magnetic beads are fully adsorbed, then carefully remove the liquid.

6. Wash 3:

Repeat step 5 once.

7. **Magnetic Separation** and Ethanol Removal: Briefly centrifuge, place the centrifuge tube on the magnetic rack and let it stand for 10 seconds to remove residual liquid (this step cannot be omitted, excessive ethanol residue may inhibit subsequent experiments); 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 difficulties in elution).

8. **Elution:** Remove the centrifuge tube, add 100μL of RNase Free Water, vortex to mix, shake at 55°C constant temperature and 1500 rpm for 5 minutes (or in a 55°C water bath, vortex mix 3-4 times during the period).

9. **Nucleic Acid Transfer:** Place the centrifuge tube on the magnetic rack, let it stand for 2 minutes to allow magnetic adsorption, transfer the RNA solution to a new centrifuge tube, and store at -80°C.

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

1. Sample Handling and Lysis

Refer to the steps outlined in the "Manual Centrifuge Tube Operation Steps".

2. Sample Loading 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	6/12
Reagent	Bead suspension（20μL）+Anhydrous ethanol（350μL）	Buffer W1A（600μL）	Buffer W2R（600μL）	Buffer W2R（600μL）	RNase Free Water（100μL）

3. Automated Extraction:

Transfer the processed supernatant (400-500μL) to the wells or slots containing ethanol and magnetic beads (corresponding to column 1/7 for the 32-channel extractor). Then, load the prepared 96-well sample plate into the nucleic acid extractor in sequence, insert the magnetic rod sleeve, open the instrument's operation program, select the appropriate program, click "run" to start the extraction.

4. Nucleic Acid Transfer:

After the instrument has completed its operation, transfer the eluate from the 6th/12th columns of the 96-well plate to clean centrifuge tubes devoid of nucleases.

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	Washing 2	0	1	60	600	3	OFF
4	4	Washing 3	0	1	60	600	3	OFF
5	6	Elution	2	5	60	100	3	60℃
6	4	Abandon beads	0	1	0	600	2	OFF

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

1. Sample Handling and Lysis

Refer to the steps outlined in the "Manual Centrifuge Tube Operation Steps".

2. 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	6
Reagent	Bead suspension（20μL）+Anhydrous ethanol（350μL）	Buffer W1A（600μL）	Buffer W2R（600μL）	Buffer W2R（600μL）	RNase Free Water（100μL）

3. Automated Extraction:

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

4. **Nucleic Acid Transfer**:

After the instrument has completed its operation, seal directly or transfer the eluate from the 6th workstation to clean centrifuge tubes devoid of nucleases, and store at -20°C for future 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
Station	1	2	3	4	6	4
Waiting time	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	2
Mixing time	00:05:00	00:02:00	00:01:00	00:01:00	00:05:00	00:00:30
Suspend	No	No	No	No	No	No
Suction time	00:01:00	00:01:00	00:01:00	00:01:00	00:01:00	00:00:00
Volume	1000μL	600μL	600μL	600μL	100μL	600μL
Temperature	—	—	—	—	60℃	—

【Basic Information】

Version Number: 1.1

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