Instruction Manual
Research Use Only

MagBeads FastRNA Kit (Ready-to-Use for MPure-32)

Magnetic bead-based Purification for total DNA from tissue and cell culture samples

Size: 96 preps
Storage: 15-25 °C
Cat. No.: 117033400
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1. Introduction to MagBeads FastRNA Kit

Magbeads FastRNA Kit is intended for simple and rapid extraction of total RNA from tissue and culture cells samples using the MPure-32™ aNAP System. The kit is based on superparamagnetic particles purification technology, no phenol-chloroform extraction or alcohol precipitation. The whole extraction process is only 60 minutes. Purified RNA is ready for downstream applications such as RT-PCR, virus RNA testing and so on. The kits buffers can be used for both manual extraction process and automatic nucleic acid extraction machines.

Magbeads FastRNA Kit combines the speed and efficiency of silica-based technology with the convenient handling of magnetic particles for purification of total RNA. Samples are lysed and RNA is purified from lysates in one step through its binding to the silica surface of the particles in the presence of a chaotropic salt. The particles are separated from the lysates using a magnet and DNA is removed by treatment with RNase-free DNase. The magnetic particles are efficiently washed, and RNA is eluted in RNase-free water.

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2. Kit Components and User Supplied Materials

2.1 MagBeads FastRNA Kit Component

MagBeads FastRNA Kit (#117033400, 96 Preps)	
Components	Package
96-Well Reagent Plates	6 plates
Proteinase K	48 mg
Protease Dissolve Buffer	5 mL
DNase I	2 x 600 µL
Buffer RTL	80 mL
Buffer MCB	15 mL
8-strip A (Cover for Magnetic Rod)	12 pieces

2.2 User Supplied Materials

- Disposable powder-free gloves.
- Pipettes (adjustable).
- Sterile pipette tips with aerosol barriers (up to 200 µl).
- Vortex mixer.
- Desktop microcentrifuge with rotor for 2 ml reaction tubes (RCF max. 16,000 x g).
- PCR box or Biological cabinet. • Vacuum aspirator with flask for removing supernatant.
- Tube racks.
- 1.5 ml polypropylene sterile tubes.
- Refrigerator for 2–8°C.
- Deep-freezer for ≤ –16°C.
- Waste bin for used tips.
- Permanent pen for labeling
- Thermostatic bath or dry block for tubes with controlled temperature and capable of incubating at 25-100°C.

3. Storage and Kit Stability

Magbeads Particles, DNase I and Proteinase K should be stored at 2 - 8°C upon arrival. However, short-term storage (up to 12 weeks) at room temperature (15 - 25°C) does not affect their performance. The remaining kit components can be stored dry at room temperature (15 - 25°C) and are stable for at least 18 months under these conditions.

4. Important Consideration Before Use

- ❑ Add 2.4 mL Protease Dissolve Buffer into Proteinase K bottle, and store at -20 ℃ after it dissolves.
- ❑ Dilute Buffer MCB with isopropanol (ratio of Buffer MCB:Isopropanol = 3 : 7) and store at room temperature.
- ❑ (Optional) 2-mercaptoethanol can be added into an aliquot of RTL Lysis Buffer before use. Add 20 µL 2-mercaptoethanol per 1 mL RTL Lysis Buffer. This mixture can be stored at room temperature for 2 weeks.

5. Safety Precautions

The user should always pay attention to the following:

• Use sterile pipette tips with aerosol filters and use new tip for every procedure.

• Store extracted positive material (samples, controls and amplicons) away from all other reagents.

• Thaw all components thoroughly at room temperature before starting an assay.

• When thawed, mix the components and centrifuge briefly.

• Use disposable gloves, laboratory coats, protect eyes while samples and reagents handling. Thoroughly wash hands afterwards.

• Do not eat, drink, smoke, apply cosmetics, or handle contact lenses in laboratory work areas.

• Do not use a kit after its expiry date.

• Dispose of all samples and unused reagents in compliance with local authorities requirements.

• Samples should be considered potentially infectious and handled in a biological cabinet in compliance with appropriate biosafety practices.

• Clean and disinfect all sample or reagent spills using a disinfectant such as 0.5% sodium hypochlorite, or other suitable disinfectant.

• Avoid contact with the skin, eyes and mucose membranes. If skin, eyes and mucose membranes contact immediately flush with water, seek medical attention.

• Material Safety Data Sheets (MSDS) are available on request.

• Use of this product should be limited to personnel trained in the techniques of DNA amplification.

• The laboratory process must be one directional; it should begin in the Extraction Area move to the Amplification and Detection Area. Do not return samples, equipment and reagents to the area in which the previous step was performed.

6. Protocol

Sample Preparation

A. Cell
Harvest cells (no more than 1 x 10^7 cells). For pelleted cells, loosen the cell pellet thoroughly by flicking the tube and add the appropriate volume of 500 µL Buffer RTL. For direct lysis of cells grown in a monolayer, add 500 µL Buffer RLT onto the cell-culture dish. Pass the lysate at least 5 times through a blunt 20-gauge needle fitted to an RNase-Free Syringe.

B. Animal Tissue
Do not use more than 20 mg of animal tissue. Disrupt and homogenize the sample, then add 600 µL Buffer RTL. After lysis, centrifuge the lysate at 14,000 x g for 3 mins at room temperature.

C. Plant Tissue
Disrupt the plant sample using liquid nitrogen. Transfer up to 50 mg of powder to 1.5 mL microcentrifuge tube and add 600 µL Buffer RTL. Vortex and centrifuge at 14,000 x g for 3 mins at room temperature.

D. Yeast Cell
Collect 5 x 10^6 yeast cells, then add 300 mg 0.4-0.6 g Glass Beads and 600 µL RTL Lysis Buffer. Vortex at maximum speed for 10 mins. Centrifuge at 10,000 x g for 3 mins at room temperature.

E. Bacterial Cell
Collect 1 x 10^8 bacterial cells, then add 300 mg 0.1-0.26 g Glass Beads and 600 µL RTL Lysis Buffer. Vortex at maximum speed for 10 mins. Centrifuge at 10,000 x g for 3 mins at room temperature.

MPure-32 Automation Purification Method

1. Transfer 500 µL of lysate and 20 µL of Proteinase K carefully to well #1 or #7 of the pre-filled reagent

2. Add 10 µL of DNase I into well #3 or #9.

3. Place the reagent plate on MPure-32™ aNAP System and run the assay with the program named “FastRNA” which has the following setting:

Step	Well	Process	Time (s)			Mixing Speed	Temp (℃)
			Mix	Wait	Attract		
1	#1/#7	Mix	60	0	60	Medium	RT
2	#2/#8	Magbeads Preparation	0	600	90	Low	RT
3	#1/#7	Bind	90	0	60	Medium	RT
4	#3/#9	DNase I Digestion*	60	900	60	Very Low	RT
5	#3/#9	Addition of Buffer MCB	60	300	60	Medium	RT
6	#4/#10	Wash 1	90	0	60	Medium	RT
7	#5/#11	Wash 2	90	0	60	Medium	RT
8	#5/#11	Dry	0	300	0	Medium	RT
9	#6/#12	Elute	300	0	120	Medium	RT
10	#1/#7	Magbeads Release	60	0	0	Medium	RT

Note: The program will pause (buzzer will sound) after the DNase I digestion step in well #3 and/or #9. At this step, press the buzzer button to stop the buzz, then add 450 µL of Buffer MCB into well #3 and/or #9. Press the pause button to continue the program to the end.

4. Transfer clear purified RNA to a clean 96-well microplate (not provided). The eluent is now ready for downstream applications. Store purified RNA at -80˚C for an extended storage.

   Note: If there are still Magnetic Beads remaining in eluent, please centrifuge at 14,000 x g for 3-5 mins and transfer the supernatant into a clean 1.5 mL microcentrifuge tube.

7. Troubleshooting

This guide may be useful in solving any problems that may arise. For further assistance, please contact our technical support team at **apac-techsupport@mpbio.com**

Problem	Recommendation
False negatives with extraction product	Degradation of the nucleic acid contained in the sample. Use a new sample, store samples appropriately.
	Loss of nucleic acid deposit. Carefully draw off the wash solution and try not to remove the nucleic acid deposit.
	Degradation of the extracted nucleic acid. Plastic free from DNAses and RNAses should be used. Use a new aliquot of kit's component.
False positives with extraction product	Contamination during sample extraction. One test tube at a time should be opened. Avoid spilling the contents of the test tube, always change tips.
	Contamination of the reagents prepared for the step. Use a new aliquot of a component.
	Contamination of the extraction zone by amplicons. Surfaces and instruments using aqueous detergents should be cleaned, wash lab coats, replace test tubes and tips in use.

8. Product Use Limitation & Warranty

The products presented in this instruction manual are for research or manufacturing use only. They are not to be used as drugs or medical devices to diagnose, cure, mitigate, treat, or prevent diseases in humans or animals, either as part of an accepted course of therapy or in experimental clinical investigation. These products are not to be used as food, food additives or general household items. Purchase of MP Biomedicals products does not grant rights to reproduce, modify, or repackage the products or any derivative thereof to third parties. MP Biomedicals makes no warranty of any kind, expressed or implied, including merchantability or fitness for any particular purpose, except that the products sold will meet our specifications at the time of delivery.

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