Instruction Manual
Research Use Only

MagBeads FastRNA Kit

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

Size: 96 preps
Storage: 15-25 °C
Cat. No.: 116572096
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1. Introduction to MagBeads FastRNA Kit

MagBeads FastDNA® Kit is intended for rapid extraction of DNA from tissue, cells, blood, saliva, swabs, blood spots, semen, and other clinical samples. DNA can be used directly for PCR, quantitative PCR, Southern Blot, detection of viral DNA and so on.

MagBeads FastDNA® Kit is based on the purification method of high binding magnetic particles. The sample is lysed and digested. DNA is released into the lysate. After addition of magnetic particles and binding solution, DNA will be adsorbed on the surface of magnetic particles, and impurities such as proteins will be removed without adsorption. The adsorbed particles were washed with washing buffer to remove the proteins and impurities, washed with ethanol to remove salts, and finally the DNA was eluted with Elution Buffer.

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2. Kit Components and User Supplied Materials

2.1 MagBeads FastRNA Kit Component

MagBeads FastRNA Kit (#116572096, 96 Preps)	
Components	Package
Magbeads RNA Particles	4.0 mL
Proteinase K	48 mg
Protease Dissolve Buffer	5 mL
DNase I	2 x 600 µL
DNase Buffer	2 x 30 mL
Buffer RTL	80 mL
Buffer MCB	30 mL
Buffer MW1	66 mL
Buffer RW2	50 mL
RNase Free Water	30 mL

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 2.5 mL Protease Dissolve Buffer into Proteinase K/Carrier RNA bottle, and store at -20 ℃ after it dissolves.
- ❑ Add 84 mL ethanol to the bottle of Buffer MW1
- ❑ Add 200 mL ethanol to the bottle of Buffer RW2
- ❑ Add 70 mL isopropanol to the bottle of Buffer MCB.
- ❑ (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.

Manual Purification Method

1. Transfer 500 µL of the lysate to a new clean 1.5 mL Tube.
2. Add 500 µL Buffer MCB, 30 µL Magbeads RNA Particles and 20 µL Proteinase K to the sample. Mix up and down 20~30 times. Stand at room temperature for 10 mins, and mix up and down for several times. Place the tube on the magnetic rack for 1 min, until the Magbeads RNA Particles have formed a tight pellet, then remove the supernatant.
3. Add 600 µL Buffer MW1 and vortex for 20s to resuspend the particles. Place the tube on the magnetic rack for 1 min, then remove the supernatant. Spin down briefly to collect liquid on the tube and remove all liquid carefully. Dry on air for 2 mins.
4. Add 300 µL DNase Mixture (290 µL DNase Buffer + 10 µL DNase I) to the sample, shake slightly to resuspend the particles and incubate at room temperature for 15 mins.
5. Add 450 µL Buffer MCB to the sample and vortex for 20s. Stand at room temperature for 5 mins and mix up and down for 2~3 times. Place the tube on the magnetic rack for 1 min, then remove the supernatant.
6. Add 600 µL Buffer MW1 and vortex for 10s to resuspend the particles. Place the tube on the magnetic rack for 1 min, then remove the supernatant.

7. Add 600 µL Buffer RW2 and vortex for 10s to resuspend the particles. Place the tube on the magnetic rack for 1 min, then remove the supernatant.
8. Repeat step 7.
9. Spin down briefly to collect the liquid on the tube, place the tube on the magnetic rack. Remove all liquid carefully. Dry at room temperature or 37℃ for 10 mins.
10. Add 30~100 µL RNase Free Water to sample, mix the particles by vortex. Stay at room temperature for 3 mins.
11. Place the tube on the magnetic rack for 3 mins. Transfer the supernatant containing the purified RNA to a new 1.5 mL centrifuge tube. Store RNA at -80°C.

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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