

MagBeads FastDNA Kit

Magnetic bead-based Purification for DNA from tissue, cells,
blood, saliva, swabs, blood spots, semen

Size: 192 preps
Storage: 15-25 °C
Cat. No.: 116575192
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1. Introduction to MagBeads FastDNA 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 FastDNA Kit Component

MagBeads FastDNA Kit (#116575192, 192 Preps)	
Components	Package
Magbeads Particles	6.6 mL
Proteinase K	90 mg
Protease Dissolve Buffer Glycerol/Tris/CaCl ₂	10 mL
RNase A Ribonuclease	40 mg
Buffer ATL Tris/EDTA/SDS	60 mL
Buffer AL Guanidine Salt	60 mL
Buffer BD Sodium perchlorate	20 mL
Buffer BW1 Guanidine Salt	110 mL
Elution Buffer Tris/EDTA	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

Proteinase K, RNase A, MagBeads Particles should be stored at 2-8°C upon arrival. However, short-term storage (up to 24 weeks) at room temperature (15-25°C) does not affect their performance. The remaining kit components can be stored at room temperature (15-25°C) and are stable for at least 18 months under these conditions.

4. Important Consideration Before Use

- Add 5 ml Protease Dissolve Buffer to the Proteinase K, and store at -20~8°C after dissolve.
- Add 2.5 ml Protease Dissolve Buffer to the RNase A, and store at -20~8°C after dissolve.
- Dilute Buffer BW1 with 140 ml 100% ethanol and store at room temperature.
- Dilute Buffer BD with 80 ml 100% ethanol and store at room temperature
- Prepare 75% Ethanol using Absolute Ethanol and store at room temperature.

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. Solid tissue (1~20mg)

1. Cut ~20mg tissue into small pieces and transfer into a new 1.5ml centrifuge tube. Add 200ul Buffer ATL and 20ul Proteinase K, shake at 55°C for 30~180 minutes.
2. (Optional) Add 10ul RNase A to the lysate and stand at room temperature for 10 min.
3. Add 200ul Buffer AL to the samples, vortex to mix and incubate at 70°C for 10 minutes.

B. Anticoagulated blood or Plasma (200uL)

1. Transfer 20ul Proteinase K to a new 1.5ml centrifuge tube.
2. Add 200ul whole blood, plasma or other body fluids to the tube, shake to mix for 5 seconds.
3. Add 200ul Buffer AL to the samples. Invert for 3~5 times, and then vortex at maximum speed for 10 seconds. Incubate at 70°C for 10 minutes.

C. Saliva sample (Preserved)

1. Transfer 20ul Proteinase K and 10ul RNase A to 1.5ml centrifuge tube.
2. Add 450ul saliva to the tube and shake to mix for 5 seconds.
3. Incubate at 55°C for 30 minutes.

D. Culture cells

1. Collect cells ($<2 \times 10^6$) by centrifuging at 2,000 x g for 5 min. Remove the supernatant.
2. Add 200ul PBS, 20ul Proteinase K and 10ul RNase A to the sample, resuspend the cells by vortexing.
3. Add 200ul Buffer AL and vortex for 10 seconds. Incubate at 70°C for 10 minutes.

E. Semen sample

1. Transfer 100ul semen to 1.5ml centrifuge tube.
2. Add 100ul Buffer ATL, 10ul DTT Solution (1M) and 20ul Proteinase K to the samples. Shake at 55°C for 30 minutes.
3. Add 200ul Buffer AL to the sample, then vortex to mix and incubate at 70°C for 10 minutes.

F. Swab DNA extraction

1. Transfer the swabs to the 2ml centrifuge tube.
2. Add ~500ul Buffer ATL and 20ul Proteinase K. Shake at 55°C for 15~30 minutes.
3. Transfer the supernatant into a new tube.

G. Blood stains/Seminal spots

1. Transfer 3 slices (3mm) of sample to the 2ml centrifuge tube. Add 250ul Buffer ATL and 20ul Proteinase K to the sample. Shake at high speed for 30~60min at 55°C.
2. Add 250ul Buffer AL to the samples, Shake at high speed for 10 min at 70°C.
3. Centrifuge at 13,000 x g for 1 min. Transfer 400ul of the supernatant to a new centrifuge tube.

H. FFPE Samples

1. Using a scalpel, trim excess paraffin off the sample block. Cut up to 1-3 sections (5-10 μm thick), put into a 1.5 ml microcentrifuge tube. Remove Paraffin by xylene or Buffer DPS (not provided).
2. Add 200 μl Buffer ATL and 20 μl Proteinase K to the sample, mix well and incubate at 56 °C for 60min, 90 °C for 60 min.
3. Cool to room temperature, add 200 μl Buffer AL and mix well.

Manual Purification Method

1. Add 20 μl Magbeads Particles and 400 μl Buffer BD to the samples or the supernatant. Mix thoroughly by inverting for 15-30 times. Incubate for 3 min with occasional inverting to mix. Place the tube on the magnetic stand for 1 minutes until the beads have formed a tight pellet. Then remove the supernatant.
2. Add 600 μl Buffer GW1 and vortex for 15 seconds to re-suspend the beads. Place the tube on the magnetic stand for 1 minute until the beads have formed a tight pellet. Then remove the supernatant.
3. Add 600 μl Buffer GW1 and vortex for 15 seconds to re-suspend the beads. Place the tube on the magnetic stand for 1 minute until the beads have form a tight pellet. Then remove the supernatant.
4. Add 600 μl 75% ethanol, and vortex for 15 seconds to re-suspend the beads. Place the tube on the magnetic stand for 1 minute until the beads have formed a tight pellet. Then remove the supernatant.
5. Add 600 μl 75% ethanol, and vortex for 15 seconds to re-suspend beads. Place the tube on the magnetic stand for 1 minute until the beads have formed a tight pellet. Then remove the supernatant.
6. Centrifuge briefly to collect the liquid on the tube. Place the tube on the magnetic stand and remove all the liquid carefully. Air dry for 10 minutes.
7. Add 50-100 μl Elution Buffer to the sample, re-suspend the beads by vortex. Incubate at 55 °C for 10 minutes with shaking. If there is no shaking device, vortex 2-3 times to mix.
8. Place the tube on the magnetic stand for 2 minutes. Transfer the supernatant containing the purified DNA to a clean 1.5ml 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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