MagBeads Fast Circulating DNA Kit (Ready-to-Use for MPure-32)

Magnetic bead-based Purification for high-quality circulating DNA (cfDNA) from cell-free body fluids (such as plasma, serum)

Size: 96 preps Storage: 15-25 °C Cat. No.: 117033900 Content Version: Feb 2024

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1. Introduction to MagBeads Fast Circulating DNA Kit

MagBeads Fast Circulating DNA Kit is intended for purification of high-quality circulating DNA (cfDNA) from cell-free body fluids (such as plasma, serum) using the MPure-32™ aNAP System. The purified DNA is suitable for direct use in downstream applications such as PCR, real-time PCR, Biochip analysis and NGS.

MagBeads Fast Circulating DNA 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

MagBeads Fast Circulating (#117033900, 96 Prep	
Components	Package
96-Well Reagent Plates	6 plates
Carrier RNA	150 µg
Proteinase K	90 mg
Protease Dissolve Buffer	5 mL
Elution Buffer	30 mL
8-strip A (Cover for Magnetic Rod)	12 pieces

2.1 MagBeads Fast Circulating DNA Kit Component

2.2 User Supplied Materials

- Disposable powder-free gloves.
- Pipettes (adjustable).
- Sterile pipette tips with aerosol barriers (up to 200 μ l).
- 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 $\leq -16^{\circ}$ 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, Carrier RNA and Magbeads Particles F should be stored at 2 - 8°C upon arrival. However, short-term storage (up to 12 weeks) at room temperature (15-25°C) will 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. The entire kit can be stored at 2-8°C, but in this case, buffers should be redissolved before use. Make sure that all buffers are at room temperature when used.

4. Important Consideration Before Use

- □ Add 4.5 mL Protease Dissolve Buffer into Proteinase K, and store at -20 °C after it dissolves.
- □ Add 0.15 mL Elution Buffer into the Carrier RNA, and store at -20°C after it dissolves.

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

MPure-32 Automation Purification Method

- 1. Peel off the sealing film on the 96-well reagent plate.
- 2. Add 30-50 µL elution buffer into well #6 and/or #12.
- 3. Transfer desired volume of sample and other components according to the following table:

	Total Sample	Well #	Components at Sample Well		
Assay Program Volume (µL)		weii#	Sample (µL)	ΡΚ (μL)	Carrier RNA (ng)
Cir_DNA_300	300	1,7	300	20	50
Cir_DNA_700	700	1,2 (sample 1); 7,8 (sample 2)	350	20	50
Cir_DNA_1000	1000	1,2,3 (sample 1); 7,8,9 (sample 2)	330	13	50

 Depending on the sample volume, run the assay with any of the following programs: Cir_DNA_300 (300 μL sample)

Step	Well	Process	Time (s)			Mixing Speed	Temp (C)
Step	Step wen		Mix	Wait	Attract	winking Speed	lenip (c)
1	#1/#7	Bind	600	0	120	Low	RT
2	#2/#8	Wash 1	150	0	60	Medium	RT
3	#3/#9	Wash 2	60	0	60	Medium	RT
4	#4/#10	Wash 3	60	0	60	Medium	RT
5	#4/#10	Dry	0	300	0	-	RT
6	#5/#11	Elute	300	0	120	Medium	RT
7	#1/#7	Magbeads Release	60	0	0	Medium	RT

Cir_DNA_700 (700 µL sample)

Chan	Well	Process		Time (s)			T (2)
Step	Step Well	Process	Mix	Wait	Attract	Mixing Speed	Temp (Ĉ)
1	#1/#7	Bind 1	300	0	120	Low	RT
2	#2/#8	Bind 2	300	0	120	Low	RT
3	#3/#9	Wash 1	90	0	60	Medium	RT
4	#4/#10	Wash 2	60	0	60	Medium	RT
5	#5/#11	Wash 3	60	0	60	Medium	RT
6	#5/#11	Dry	0	300	0	-	RT
7	#6/#12	Elute	300	0	120	Medium	RT
8	#1/#7	Magbeads Release	60	0	0	Medium	RT

Cir_DNA_1000 (1000 µL sample)

Cham	Well	Process		Time (s)		Mining Speed	Temp (Ĉ)
Step	Step Well	Process	Mix	Wait	Attract	Mixing Speed	
1	#1/#7	Bind 1	300	0	120	Low	RT
2	#2/#8	Bind 2	300	0	120	Low	RT
3	#3/#9	Bind 3	300	0	120	Low	RT
4	#4/#10	Wash 1	90	0	60	Medium	RT
5	#5/#11	Wash 2	60	0	60	Medium	RT
6	#5/#11	Dry	0	300	0	-	RT
7	#6/#12	Elute	300	0	120	Medium	RT
8	#1/#7	Magbeads Release	60	0	0	Medium	RT

5. Transfer the eluted nucleic acid into a clean 1.5 mL microcentrifuge tube. The

eluent is now ready for downstream applications. Store the purified nucleic acid at -20° C for an extended storage.

Note: If there are still Magnetic Beads remaining in eluted DNA, please centrifuge at $14,000 \times g$ 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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